

FUSED HETEROCYCLIC ISOXAZOLINE DERIVATIVES
AND THEIR USE AS ANTI-DEPRESSANTS

The invention concerns fused heterocyclic isoxazoline derivatives, more in particular tetrahydropyranoisoxazole, hexahydroisoxazolopyridine, tetrahydrothiopyranoisoxazole and hexahydrobenzoisoxazole derivatives fused to a heterocyclic ring system via the 6-membered ring of the bicyclic moiety as well as processes for their preparation, pharmaceutical compositions comprising them and their use as a medicine, in particular for treating depression, anxiety, movement disorders, psychosis, Parkinson's disease and body weight disorders including anorexia nervosa and bulimia.

The invention also relates to novel combination of said fused heterocyclic isoxazoline derivatives with antidepressants, anxiolytics, antipsychotics and anti-Parkinson's disease drugs.

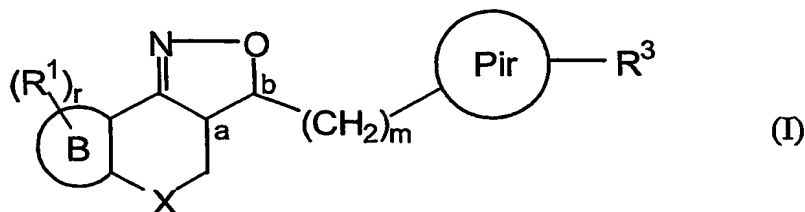
Tetrahydronaphtalene and indane derivatives showing anti-depressant activity are known from EP-361 577 B1. These compounds are typical monoamine reuptake blockers with additional α_2 -adrenoceptor antagonist activity and they show anti-depressant activity without being sedative.

The problems associated with the compounds according to the state of the art is that the compounds cause considerable side-effects, such as nausea, excitation, an increased heart rate and a reduced sexual function. Furthermore, it requires a long time, in particular 3-4 weeks, before the response starts.

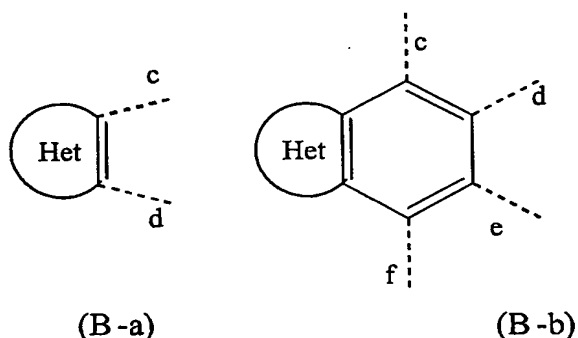
The purpose of the present invention is to provide novel compounds for treating depression, anxiety, movement disorders, psychosis, schizophrenia and body weight disorders, in particular compounds that do not exhibit the aforementioned disadvantages.

-2-

- 5 The present invention relates to novel isoxazoline derivatives according to the general Formula (I)



- the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof, wherein :
- 10 X is CH₂, N-R⁷, S or O ;
- R⁷ is selected from the group of hydrogen, alkyl, Ar, Ar-alkyl, alkylcarbonyl, alkyloxycarbonyl and mono- and dialkylaminocarbonyl ;
- B is a radical, optionally substituted with *r* radicals R¹, according to anyone of
- 15 Formula (B-a) or (B-b) and fused to the isoxazolinyl moiety by either of the bond pairs (c,d), (d,e) or (e,f)



wherein

- 20 Het is an optionally substituted 5- or 6-membered heterocyclic ring, selected from the group of pyridinyl, pyrazinyl, pyrimidinyl, pyridazinyl, furanyl, thienyl, pyrrolyl, oxazolyl, thiazolyl, imidazolyl, pyrazolyl, isothiazolyl, isoxazolyl, oxadiazolyl and triazolyl ;
- each R¹ is, independently from each other, selected from the group of hydrogen,
- 25 hydroxy, amino, nitro, cyano, halo and alkyl and, only when R¹ is attached to a *N*-atom, is further selected from the group of alkyloxyalkyl, alkyloxyalkoxyalkyl, alkyloxycarbonylalkyl, formyl, alkylcarbonyl, alkyloxycarbonyl, alkyloxyalkylcarbonyl and mono- and dialkylamino-carbonyl ;
- 30 *r* is an integer ranging from 0 to 6 ;

-3-

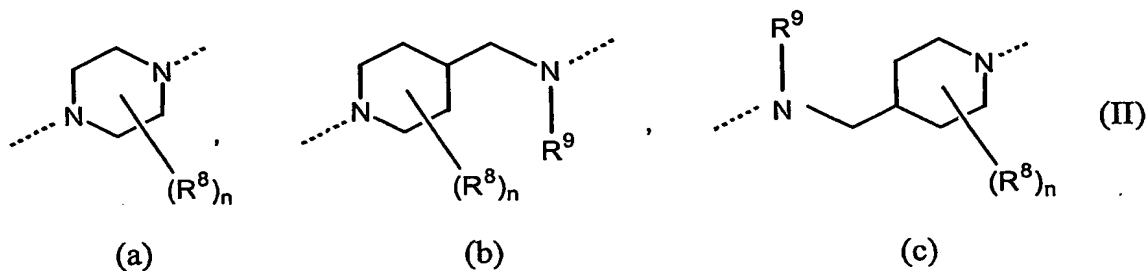
5

a and b are asymmetric centers ;

$(CH_2)_m$ is a straight hydrocarbon chain of m carbon atoms, m being an integer ranging from 1 to 4 ;

Pir is a radical according to any one of Formula (IIa), (IIb) or (IIc)

10



optionally substituted with n radicals R^8 , wherein :

each R^8 is independently from each other, selected from the group of hydroxy, amino, nitro, cyano, halo and alkyl ;

15 n is an integer ranging from 0 to 5 ;

R^9 is selected from the group of hydrogen, alkyl and formyl ;

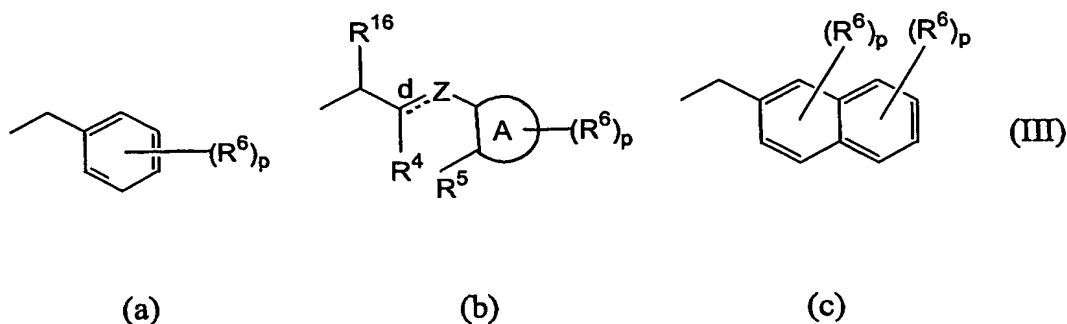
R^3 represents an optionally substituted aromatic homocyclic or heterocyclic ring system together with an optionally substituted and partially or completely hydrogenated hydrocarbon chain of 1 to 6 atoms long with which said ring system is attached to the Pir radical and of which may contain one or more heteroatoms selected from the group of O, N and S ; and

20

Ar is phenyl or naphthyl, optionally substituted with one or more halo, cyano, oxo, hydroxy, alkyl, formyl, alkyloxy or amino radicals.

25 More in particular, the invention relates to compounds according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the N-oxide form thereof, wherein R^3 is a radical according to any one of Formula (IIIa), (IIIb) or (IIIc)

-4-



wherein :

d is a single bond while Z is a bivalent radical selected from the group of $-\text{CH}_2-$, $-\text{C}(=\text{O})-$, $-\text{CH}(\text{OH})-$, $-\text{C}(=\text{N}-\text{OH})-$, $-\text{CH}(\text{alkyl})-$, $-\text{O}-$, $-\text{S}-$, $-\text{S}(=\text{O})-$, $-\text{NH}-$ and $-\text{SH}-$; or d is a double bond while Z is a trivalent radical of formula $=\text{CH}-$ or $=\text{C}(\text{alkyl})-$;

A is a 5- or 6-membered aromatic homocyclic or heterocyclic ring, selected from the group of phenyl, pyranyl, pyridinyl, pyrazinyl, pyrimidinyl, pyridazinyl, thienyl, isothiazolyl, pyrrolyl, imidazolyl, pyrazolyl, furanyl, oxadiazolyl and isoxazolyl ;

p is an integer ranging from 0 to 6 ;

R^4 and R^5 are each, independently from each other, selected from the group of hydrogen, alkyl, Ar, biphenyl, halo and cyano ; or

R^4 and R^5 may be taken together to form a bivalent radical $-\text{R}^4-\text{R}^5-$ selected from the group of $-\text{CH}_2-$, $=\text{CH}-$, $-\text{CH}_2-\text{CH}_2-$, $-\text{CH}=\text{CH}-$, $-\text{O}-$, $-\text{NH}-$, $=\text{N}-$, $-\text{S}-$, $-\text{CH}_2\text{N}(-\text{alkyl})-$, $-\text{N}(-\text{alkyl})\text{CH}_2-$, $-\text{CH}_2\text{NH}-$, $-\text{NHCH}_2-$, $-\text{CH}=\text{N}-$, $-\text{N}=\text{CH}-$, $-\text{CH}_2\text{O}-$ and $-\text{OCH}_2-$;

each R^6 is independently from each other, selected from the group of hydroxy, amino, nitro, cyano, halo, carboxyl, alkyl, Ar, alkyloxy, Ar-oxy, alkyl-carbonyloxy, alkyloxycarbonyl, alkylthio, mono- and di(alkyl)amino, alkylcarbonylamino, mono- and di(alkyl)aminocarbonyl, mono- and di(alkyl)aminocarbonyloxy, mono- and di(alkyl)aminoalkyloxy ; or

two vicinal radicals R^6 may be taken together to form a bivalent radical $-\text{R}^6-\text{R}^6-$ selected from the group of $-\text{CH}_2-\text{CH}_2-\text{O}-$, $-\text{O}-\text{CH}_2-\text{CH}_2-$, $-\text{O}-\text{CH}_2-\text{C}(=\text{O})-$, $-\text{C}(=\text{O})-\text{CH}_2-\text{O}-$, $-\text{O}-\text{CH}_2-\text{O}-$, $-\text{CH}_2-\text{O}-\text{CH}_2-$, $-\text{O}-\text{CH}_2-\text{CH}_2-\text{O}-$, $-\text{CH}=\text{CH}-\text{CH}=\text{CH}-$, $-\text{CH}=\text{CH}-\text{CH}=\text{N}-$, $-\text{CH}=\text{CH}-\text{N}=\text{CH}-$, $-\text{CH}=\text{N}-\text{CH}=\text{CH}-$, $-\text{N}=\text{CH}-\text{CH}=\text{CH}-$, $-\text{CH}_2-\text{CH}_2-\text{CH}_2-$, $-\text{CH}_2-\text{CH}_2-\text{C}(=\text{O})-$, $-\text{C}(=\text{O})-\text{CH}_2-\text{CH}_2-$, $-\text{CH}_2-\text{C}(=\text{O})-\text{CH}_2-$ and $-\text{CH}_2-\text{CH}_2-\text{CH}_2-\text{CH}_2-$ and

R^{16} is selected from the group of hydrogen, alkyl, Ar and Ar-alkyl.

-5-

5 Preferably, the invention relates to those compounds according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof, wherein $X = O$; $m = 1$; B is a radical according to Formula (B-a) or (B-b), Pir is a radical according to Formula (IIa) wherein $n = 0$; R^3 is a radical according to any one of Formula (IIIa),
10 (IIIb) or (IIIc) wherein d is a double bond while Z is a trivalent radical of formula $=CH-$ or $=C(alkyl)-$; A is a phenyl ring ; R^4 is hydrogen or alkyl ; R^5 and R^{16} are each hydrogen ; R^6 is hydrogen or halo and $p = 1$.

15 Preferably, the invention relates to those compounds according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof, wherein Het is selected from the group of pyridinyl, thienyl and pyrrolyl, each radical optionally substituted on a N atom with a radical selected from the group of hydrogen, alkyl, alkyloxyalkyloxyalkyl, alkyloxycarbonylalkyl, alkylcarbonyl, alkyloxycarbonyl and alkyloxyalkylcarbonyl.

20 In the framework of this application, alkyl defines straight or branched saturated hydrocarbon radicals having from 1 to 6 carbon atoms, for example methyl, ethyl, propyl, butyl, 1-methylpropyl, 1,1-dimethylethyl, pentyl, hexyl ; or alkyl defines cyclic saturated hydrocarbon radicals having from 3 to 6 carbon atoms, for example
25 cyclopropyl, methylcyclopropyl, cyclobutyl, cyclopentyl and cyclohexyl. The definition of alkyl also comprises alkyl radicals that are substituted with one or more halo, cyano, oxo, hydroxy, formyl or amino radicals, for example hydroxyalkyl, in particular hydroxymethyl and hydroxyethyl and polyhaloalkyl, in particular difluoromethyl and trifluoromethyl.

30 In the framework of this application, Ar is phenyl or naphthyl, optionally substituted with one or more halo, cyano, oxo, hydroxy, alkyl, formyl, alkyloxy or amino radicals, such as for example, 3-fluoro-phenyl or 3-fluoro-naphthyl.

35 In the framework of this application, halo is generic to fluoro, chloro, bromo and iodo.

The pharmaceutically acceptable salts are defined to comprise the therapeutically active non-toxic acid addition salts forms that the compounds according to Formula (I) are able to form. Said salts can be obtained by treating the base form of the compounds
40 according to Formula (I) with appropriate acids, for example inorganic acids, for example hydrohalic acid, in particular hydrochloric acid, hydrobromic acid, sulfuric acid, nitric acid and phosphoric acid ; organic acids, for example acetic acid,

-6-

- 5 hydroxyacetic acid, propanoic acid, lactic acid, pyruvic acid, oxalic acid, malonic acid, succinic acid, maleic acid, fumaric acid, malic acid, tartaric acid, citric acid, methanesulfonic acid, ethanesulfonic acid, benzenesulfonic acid, p-toluenesulfonic acid, cyclamic acid, salicyclic acid, p-aminosalicylic acid and pamoic acid.
- 10 The compounds according to Formula (I) containing acidic protons may also be converted into their therapeutically active non-toxic metal or amine addition salts forms by treatment with appropriate organic and inorganic bases. Appropriate base salts forms comprise, for example, the ammonium salts, the alkaline and earth alkaline metal salts, in particular lithium, sodium, potassium, magnesium and calcium salts, salts with
- 15 organic bases, e.g. the benzathine, *N*-methyl-D-glucamine, hybramine salts, and salts with amino acids, for example arginine and lysine.

Conversely, said salts forms can be converted into the free forms by treatment with an appropriate base or acid.

20

The term addition salt as used in the framework of this application also comprises the solvates that the compounds according to Formula (I) as well as the salts thereof, are able to form. Such solvates are, for example, hydrates and alcoholates.

- 25 The *N*-oxide forms of the compounds according to Formula (I) are meant to comprise those compounds of Formula (I) wherein one or several nitrogen atoms are oxidized to the so-called *N*-oxide, particularly those *N*-oxides wherein one or more nitrogens of the piperaziny radical are *N*-oxidized.

- 30 The term "stereochemically isomeric forms" as used hereinbefore defines all the possible isomeric forms that the compounds of Formula (I) may possess. Unless otherwise mentioned or indicated, the chemical designation of compounds denotes the mixture of all possible stereochemically isomeric forms, said mixtures containing all diastereomers and enantiomers of the basic molecular structure. More in particular,
- 35 stereogenic centers may have the *R*- or *S*-configuration; substituents on bivalent cyclic (partially) saturated radicals may have either the *cis*- or *trans*-configuration. Compounds encompassing double bonds can have an *E* or *Z*-stereochemistry at said double bond. Stereochemically isomeric forms of the compounds of Formula (I) are obviously intended to be embraced within the scope of this invention.

40

Following CAS nomenclature conventions, when two stereogenic centers of known absolute configuration are present in a molecule, an *R* or *S* descriptor is assigned (based

-7-

5 on Cahn-Ingold-Prelog sequence rule) to the lowest-numbered chiral center, the reference center. The configuration of the second stereogenic center is indicated using relative descriptors $[R^*,R^*]$ or $[R^*,S^*]$, where R^* is always specified as the reference center and $[R^*,R^*]$ indicates centers with the same chirality and $[R^*,S^*]$ indicates centers of unlike chirality. For example, if the lowest-numbered chiral center in the molecule has an *S* configuration and the second center is *R*, the stereo descriptor would be specified as *S*- $[R^*,S^*]$. If “ α ” and “ β ” are used : the position of the highest priority substituent on the asymmetric carbon atom in the ring system having the lowest ring number, is arbitrarily always in the “ α ” position of the mean plane determined by the ring system. The position of the highest priority substituent on the other asymmetric carbon atom in the ring system (hydrogen atom in compounds according to Formula (I)) relative to the position of the highest priority substituent on the reference atom is denominated “ α ”, if it is on the same side of the mean plane determined by the ring system, or “ β ”, if it is on the other side of the mean plane determined by the ring system.

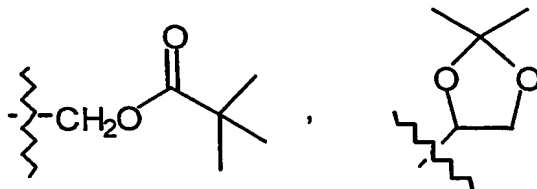
20 Compounds according to Formula (I) and some of the intermediate compounds have at least two stereogenic centers in their structure, respectively denoted a and b in Formula (I). Due to the synthetic pathway followed for the synthesis of the tricyclic system, the configuration of those two asymmetric centers a and b is predetermined, so that the relative configuration of center a is S^* and of center b is R^* .

The invention also comprises derivative compounds (usually called “pro-drugs”) of the pharmacologically-active compounds according to the invention, which are degraded *in vivo* to yield the compounds according to the invention. Pro-drugs are usually (but not always) of lower potency at the target receptor than the compounds to which they are degraded. Pro-drugs are particularly useful when the desired compound has chemical or physical properties that make its administration difficult or inefficient. For example, the desired compound may be only poorly soluble, it may be poorly transported across the mucosal epithelium, or it may have an undesirably short plasma half-life. Further discussion on pro-drugs may be found in Stella, V. J. *et al.*, “Prodrugs”, *Drug Delivery Systems*, 1985, pp. 112-176, and *Drugs*, 1985, 29, pp. 455-473.

40 Pro-drugs forms of the pharmacologically-active compounds according to the invention will generally be compounds according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof, having an acid group which is esterified or amidated. Included

-8-

- 5 in such esterified acid groups are groups of the formula $-\text{COOR}^x$, where R^x is a C_{1-6} alkyl, phenyl, benzyl or one of the following groups :



- 10 Amidated groups include groups of the formula $-\text{CONR}^y\text{R}^z$, wherein R^y is H, C_{1-6} alkyl, phenyl or benzyl and R^z is $-\text{OH}$, H, C_{1-6} alkyl, phenyl or benzyl.

Compounds according to the invention having an amino group may be derivatised with a ketone or an aldehyde such as formaldehyde to form a Mannich base. This base will hydrolyze with first order kinetics in aqueous solution.

- 15 The compounds of Formula (I) as prepared in the processes described below may be synthesized in the form of racemic mixtures of enantiomers that can be separated from one another following art-known resolution procedures. The racemic compounds of Formula (I) may be converted into the corresponding diastereomeric salt forms by reaction with a suitable chiral acid. Said diastereomeric salt forms are subsequently
- 20 separated, for example, by selective or fractional crystallization and the enantiomers are liberated therefrom by alkali. An alternative manner of separating the enantiomeric forms of the compounds of Formula (I) involves liquid chromatography using a chiral stationary phase. Said pure stereochemically isomeric forms may also be derived from the corresponding pure stereochemically isomeric forms of the appropriate starting
- 25 materials, provided that the reaction occurs stereospecifically. Preferably if a specific stereoisomer is desired, said compound would be synthesized by stereospecific methods of preparation. These methods will advantageously employ enantiomerically pure starting materials.

- 30 The compounds according to the invention, in particular compounds according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof, have surprisingly been shown to have selective serotonin (5-HT) reuptake inhibitor activity in combination with additional α_2 -adrenoceptor antagonist activity and show a strong
- 35 anti-depressant and/or anxiolytic activity and/or antipsychotic and/or a body weight control activity without being sedative. Also, in view of their selective serotonin (5-HT) reuptake inhibitor as well as α_2 -adrenoceptor antagonist activity, compounds according to the invention are also suitable for treatment and/or prophylaxis in diseases

-9-

5 where either one of the activities alone or the combination of said activities may be of therapeutic use. In particular, the compounds according to the invention may be suitable for treatment and/or prophylaxis in the following diseases :

- Central nervous system disorders, including :

- Mood disorders, including particularly major depressive disorder, depression
10 with or without psychotic features, catatonic features, melancholic features, atypical features of postpartum onset and, in the case of recurrent episodes, with or without seasonal pattern, dysthymic disorder, bipolar I disorder, bipolar II disorder, cyclothymic disorder, recurrent brief depressive disorder, mixed affective disorder, bipolar disorder not otherwise specified, mood disorder due
15 to a general medical condition, substance-induced mood disorder, mood disorder not otherwise specified, seasonal affective disorder and premenstrual dysphoric disorders.
- Anxiety disorders, including panic attack, agoraphobia, panic disorder without agoraphobia, agoraphobia without history of panic disorder, specific phobia,
20 social phobia, obsessive-compulsive disorder, posttraumatic stress disorder, acute stress disorder, generalized anxiety disorder, anxiety disorder due to a general medical condition, substance-induced anxiety disorder and anxiety disorder not otherwise specified.
- Stress-related disorders associated with depression and/or anxiety, including
25 acute stress reaction, adjustment disorders (brief depressive reaction, prolonged depressive reaction, mixed anxiety and depressive reaction, adjustment disorder with predominant disturbance of other emotions, adjustment disorder with predominant disturbance of conduct, adjustment disorder with mixed disturbance of emotions and conduct, adjustment disorders with other specified
30 predominant symptoms) and other reactions to severe stress.
- Dementia, amnesic disorders and cognitive disorders not otherwise specified, especially dementia caused by degenerative disorders, lesions, trauma, infections, vascular disorders, toxins, anoxia, vitamin deficiency or endocrinic disorders, or amnesic disorders caused by alcohol or other causes of thiamin
35 deficiency, bilateral temporal lobe damage due to Herpes simplex encephalitis and other limbic encephalitis, neuronal loss secondary to anoxia / hypoglycemia / severe convulsions and surgery, degenerative disorders, vascular disorders or pathology around ventricle III.
- Cognitive disorders due to cognitive impairment resulting from other medical
40 conditions.
- Personality disorders, including paranoid personality disorder, schizoid personality disorder, schizotypal personality disorder, antisocial personality

-10-

- 5 disorder, borderline personality disorder, histrionic personality disorder, narcissistic personality disorder, avoidant personality disorder, dependent personality disorder, obsessive-compulsive personality disorder and personality disorder not otherwise specified.
- 10 • Schizoaffective disorders resulting from various causes, including schizoaffective disorders of the manic type, of the depressive type, of mixed type, paranoid, disorganized, catatonic, undifferentiated and residual schizophrenia, schizophreniform disorder, schizoaffective disorder, delusional disorder, brief psychotic disorder, shared psychotic disorder, substance-induced psychotic disorder and psychotic disorder not otherwise specified.
- 15 • Akinesia, akinetic-rigid syndromes, dyskinesia and medication-induced parkinsonism, Gilles de la Tourette syndrome and its symptoms, tremor, chorea, myoclonus, tics and dystonia.
- Attention-deficit / hyperactivity disorder (ADHD).
- 20 • Parkinson's disease, drug-induced Parkinsonism, post-encephalitic Parkinsonism, progressive supranuclear palsy, multiple system atrophy, corticobasal degeneration, parkinsonism-ALS dementia complex and basal ganglia calcification.
- Dementia of the Alzheimer's type, with early or late onset, with depressed mood.
- 25 • Behavioral disturbances and conduct disorders in dementia and the mentally retarded, including restlessness and agitation.
- Extra-pyramidal movement disorders.
- Down's syndrome.
- Akathisia.
- 30 • Eating Disorders, including anorexia nervosa, atypical anorexia nervosa, bulimia nervosa, atypical bulimia nervosa, overeating associated with other psychological disturbances, vomiting associated with other psychological disturbances and non-specified eating disorders.
- AIDS-associated dementia.
- 35 • Chronic pain conditions, including neuropathic pain, inflammatory pain, cancer pain and post-operative pain following surgery, including dental surgery. These indications might also include acute pain, skeletal muscle pain, low back pain, upper extremity pain, fibromyalgia and myofascial pain syndromes, orofascial pain, abdominal pain, phantom pain, tic douloureux and atypical face pain, nerve root damage and arachnoiditis, geriatric pain, central pain and inflammatory pain.
- 40 • Neurodegenerative diseases, including Alzheimer's disease, Huntington's chorea, Creutzfeld-Jacob disease, Pick's disease, demyelinating disorders, such as multiple

-11-

5 sclerosis and ALS, other neuropathies and neuralgia, multiple sclerosis, amyotropical lateral sclerosis, stroke and head trauma.

- Addiction disorders, including :

- Substance dependence or abuse with or without physiological dependence, particularly where the substance is alcohol, amphetamines, amphetamine-like
10 substances, caffeine, cannabis, cocaine, hallucinogens, inhalants, nicotine, opioids, phencyclidine, phencyclidine-like compounds, sedative-hypnotics, benzodiazepines and/or other substances, particularly useful for treating withdrawal from the above substances and alcohol withdrawal delirium.
- Mood disorders induced particularly by alcohol, amphetamines, caffeine,
15 cannabis, cocaine, hallucinogens, inhalants, nicotine, opioids, phencyclidine, sedatives, hypnotics, anxiolitics and other substances.
- Anxiety disorders induced particularly by alcohol, amphetamines, caffeine, cannabis, cocaine, hallucinogens, inhalants, nicotine, opioids, phencyclidine, sedatives, hypnotics, anxiolitics and other substances and adjustment disorders
20 with anxiety.

- Smoking cessation.

- Body weight control, including obesity.

- Sleep disorders and disturbances, including :

- Dyssomnias and/or parasomnias as primary sleep disorders, sleep disorders
25 related to another mental disorder, sleep disorder due to a general medical condition and substance-induced sleep disorder.
- Circadian rhythms disorders.
- Improving the quality of sleep.

- Sexual dysfunction, including sexual desire disorders, sexual arousal disorders,
30 orgasmic disorders, sexual pain disorders, sexual dysfunction due to a general medical condition, substance-induced sexual dysfunction and sexual dysfunction not otherwise specified.

35 The present invention thus also relates to compounds according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof, the *N*-oxide form thereof, as well as the prodrugs thereof for use as a medicine, in particular for the treatment and/or prophylaxis of depression, anxiety, movement disorders, psychosis, Parkinson's disease and body weight disorders.

40 The present invention also relates to a method for the treatment and/or prophylaxis of diseases where either one of the activities (selective serotonin (5-HT) reuptake

-12-

inhibitor and α_2 -adrenoceptor antagonist activity) alone or the combination of said activities may be of therapeutic use, in particular for the treatment and/or prophylaxis of depression, anxiety, movement disorders, psychosis, Parkinson's disease and body weight disorders comprising administering to a human in need of such administration an affective amount of a compound according to the invention, in particular according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof, the *N*-oxide form thereof, as well as the prodrugs thereof.

The invention also relates to a pharmaceutical composition comprising a pharmaceutically acceptable carrier and, as active ingredient, a therapeutically effective amount of a compound according to the invention, in particular a compound according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof or a prodrug as defined above.

The compounds according to the invention, in particular the compounds according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof and the prodrugs, or any subgroup thereof may be formulated into various pharmaceutical forms for administration purposes. As appropriate compositions there may be cited all compositions usually employed for systemically administering drugs. To prepare the pharmaceutical compositions of this invention, an effective amount of the particular compound, optionally in addition salt form, as the active ingredient is combined in intimate admixture with a pharmaceutically acceptable carrier, which carrier may take a wide variety of forms depending on the form of preparation desired for administration. These pharmaceutical compositions are desirable in unitary dosage form suitable, in particular, for administration orally, rectally, percutaneously, by parenteral injection or by inhalation. For example, in preparing the compositions in oral dosage form, any of the usual pharmaceutical media may be employed such as, for example, water, glycols, oils, alcohols and the like in the case of oral liquid preparations such as suspensions, syrups, elixirs, emulsions and solutions; or solid carriers such as starches, sugars, kaolin, diluents, lubricants, binders, disintegrating agents and the like in the case of powders, pills, capsules and tablets. Because of their ease in administration, tablets and capsules represent the most advantageous oral dosage unit forms in which case solid pharmaceutical carriers are obviously employed. For parenteral compositions, the carrier will usually comprise sterile water, at least in large part, though other ingredients, for example, to aid solubility, may be included. Injectable solutions, for

-13-

5 example, may be prepared in which the carrier comprises saline solution, glucose solution or a mixture of saline and glucose solution. Injectable suspensions may also be prepared in which case appropriate liquid carriers, suspending agents and the like may be employed. Also included are solid form preparations that are intended to be converted, shortly before use, to liquid form preparations. In the compositions suitable
10 for percutaneous administration, the carrier optionally comprises a penetration enhancing agent and/or a suitable wetting agent, optionally combined with suitable additives of any nature in minor proportions, which additives do not introduce a significant deleterious effect on the skin. Said additives may facilitate the administration to the skin and/or may be helpful for preparing the desired compositions.
15 These compositions may be administered in various ways, e.g., as a transdermal patch, as a spot-on, as an ointment.

It is especially advantageous to formulate the aforementioned pharmaceutical compositions in unit dosage form for ease of administration and uniformity of dosage.
20 Unit dosage form as used herein refers to physically discrete units suitable as unitary dosages, each unit containing a predetermined quantity of active ingredient calculated to produce the desired therapeutic effect in association with the required pharmaceutical carrier. Examples of such unit dosage forms are tablets (including scored or coated tablets), capsules, pills, powder packets, wafers, suppositories,
25 injectable solutions or suspensions and the like, and segregated multiples thereof.

The compounds according to the invention may also be suitable as add-on treatment and/or prophylaxis in the above listed diseases in combination with any combination of compounds selected from the group of antidepressants, anxiolytics, antipsychotics
30 and/or anti-Parkinson's disease drugs which are currently available or in development or which will become available in the future, to improve efficacy and/or onset of action. This is evaluated in rodent models in which antidepressants, anxiolytics, antipsychotics and/or anti-Parkinson's disease drugs are shown to be active. For example, compounds are evaluated in combination with antidepressants, anxiolytics,
35 antipsychotics and/or anti-Parkinson's disease drugs for attenuation of stress-induced hyperthermia.

The invention therefore also relates to a pharmaceutical composition comprising the compounds according to the invention, in particular the compounds according to
40 Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof, and the prodrugs and one or more other compounds selected from the group of antidepressants,

-14-

5 anxiolytics, antipsychotics and anti-Parkinson's disease drugs.

The invention also relates to the use of a pharmaceutical composition according to the invention for the manufacture of a medicament to improve efficacy and/or onset of action in the treatment and/or prophylaxis of depression, anxiety, movement disorders, psychosis, Parkinson's disease and body weight disorders.

Further, the invention relates to the use of a compound according to the invention for the manufacture of a medicament for the treatment and/or prophylaxis of depression, anxiety, movement disorders, psychosis, Parkinson's disease and body weight disorders, said treatment comprising the simultaneous or sequential administration of a compound according to the invention and one or more other compounds selected from the group of antidepressants, anxiolytics, anti-psychosis and anti-Parkinson's drugs. The invention further relates to a process for making a pharmaceutical composition comprising mixing a compound according to the invention, in particular the compounds according to Formula (I), the pharmaceutically acceptable acid or base addition salts thereof, the stereochemically isomeric forms thereof and the *N*-oxide form thereof, and the prodrugs, or any subgroup thereof and a compound selected from the group of antidepressants, anxiolytics, antipsychotics and anti-Parkinson's disease drugs and a pharmaceutically acceptable carrier.

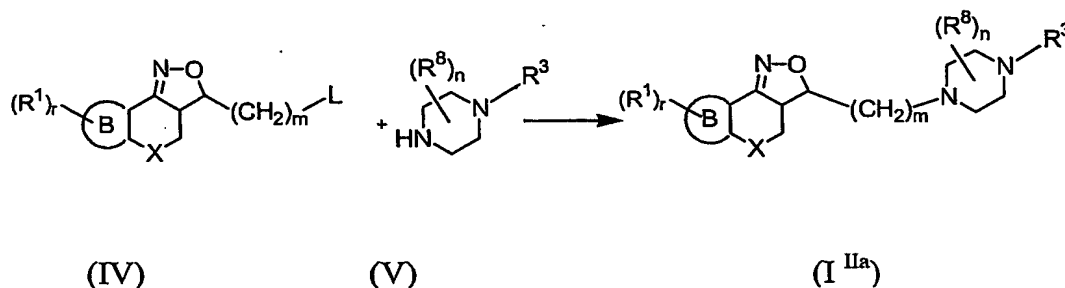
In vitro receptor and neurotransmitter transporter binding and signal-transduction studies can be used to evaluate the α_2 -adrenoceptor antagonism activity and serotonin (5-HT) reuptake inhibitor activity of the present compounds. As indices for central penetration and potency to block the α_2 -adrenoceptors and serotonin transporters, respectively, *ex vivo* α_2 -adrenoceptor and serotonin transporter occupancy can be used. As indices of α_2 -adrenoceptor antagonism *in vivo*, the reversal of the loss of righting reflex, observed in rats after subcutaneous injection or oral dosage of the compound before intravenous medetomidine administration in rats can be used (medetomidine-test). As indices of serotonin (5-HT) reuptake inhibition activity, the inhibition of head-twitches and excitation in rats, observed after subcutaneous injection or oral dosage of the compound before subcutaneous p-chloroamphetamine administration in rats can be used (pCA-test).

The compounds according to the invention can generally be prepared by a succession of steps, each of which is known to the skilled person.

In particular, the compounds according to Formula (I) with a Pir-radical according to

-15-

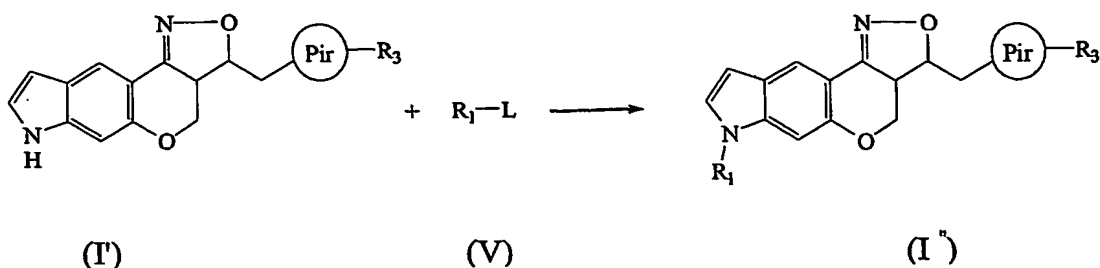
5 Formula (IIa), (IIb) or (IIc) can be prepared by a nucleophilic substitution reaction with a substituted piperazine according to Formula (V) on an intermediate compound of Formula (IV). These reactions may be carried out in a reaction inert solvent such as 1,4-dioxane, methylisobutylketone (MIBK), acetonitrile or *N,N*-dimethylformamide, in the presence of a suitable base such as NaHCO₃, Na₂CO₃, piperidine or
 10 triethylamine, or even without a base, using in this latter case excess of reagent of Formula (V). Convenient reaction temperatures range between 100°C and 150°C.



In compound (IV), L represents any suitable reactive leaving group, in particular halo, such as chloro, bromo or iodo or sulfonyloxy, such as 4-methylsulphonyloxy or
 15 4-methylbenzenesulphonyloxy.

The compounds according to the invention can easily be converted into each other. In case the B-radical is an *N*-containing heterocyclic radical, such as e.g. indol, the
 20 nitrogen atom of such a final compound according to Formula (I') can be alkylated or acylated according to art-known procedures to give final compounds of Formula (I''). Alkylation reactions can be carried out in the presence of the corresponding alkylating agent, for example any haloalkyl compound, in the presence of a base, such as NaOH, KOH, Na₂CO₃, K₂CO₃ or a mixture thereof, and an inert solvent, for example
 25 acetonitrile, tetrahydrofuran or a mixture thereof. Acylation reactions can be carried out in the presence of an acylating agent, for example acylhalides, isocyanates or acid anhydrides ; a strong base, such as NaOH, KOH or BEMP (2-tert-butylimino-2-diethylamino-1,3-dimethylperhydro-1,3,2-diaza-phosphorine) either or not supported in an inert polymer, such as polystyrene; and an inert solvent, for example
 30 dichloromethane or tetrahydrofuran. L is a leaving group, in particular halo such as chloro, bromo, iodo ; or sulphonyloxy or 4-methylsulphonyloxy ; and R¹ is any alkyl or acyl group.

-16-

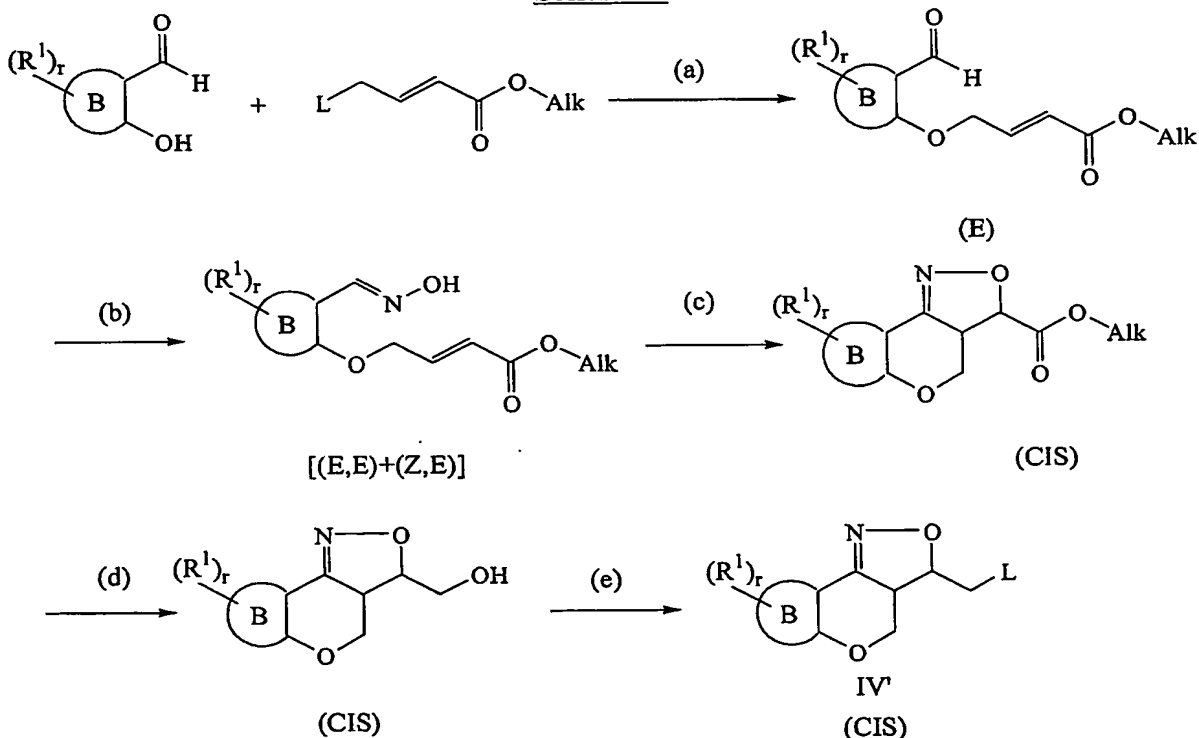


5

The starting materials and some of the intermediate compounds are compounds that are either commercially available or may be prepared according to conventional reaction procedures generally known in the art. For example, intermediate compounds of Formula (IV') in which X=O may be prepared according to the following reaction scheme 1.

10

Scheme 1



15

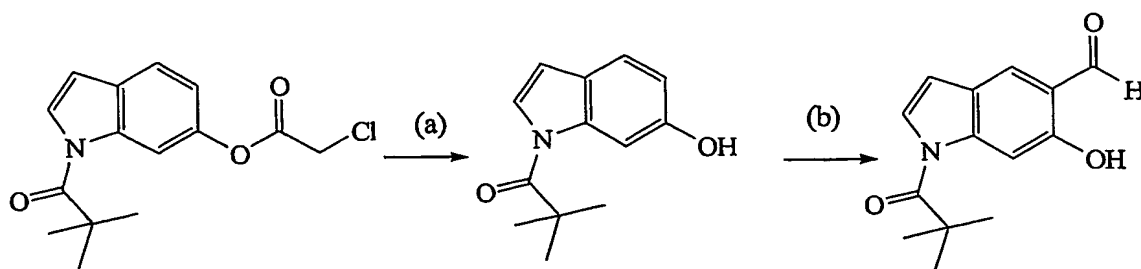
An intermediate compound of Formula (IV') can be prepared from hydroxyaldehydes by reaction with commercially available crotonates in the presence of a base, such as K_2CO_3 , Na_2CO_3 or NaH in an inert solvent, such as 2-propanone or dimethylformamide (step a). The resulting intermediate compound is converted into the oxime in a temperature range of $-10^\circ C$ to $0^\circ C$ using art-known techniques, such as hydroxylamine hydrochloride in the presence of a suitable base, such as $AcONa$, $NaHCO_3$ or pyridine in a reaction inert solvent, for example ethanol (step b). The resulting oxime-

20

-17-

intermediate compound is further oxidized into its nitrile oxide and the subsequent in situ intramolecular cycloaddition yields an isoxazoline compound (step c). The oxidation can be carried out using sodium hypochlorite solution in the presence of triethylamine in an inert solvent, such dichloromethane at room temperature. Oxidation can also be performed using chloramine-T hydrate (*N*-chloro-4-methylbenzene-sulphonamide, sodium salt), by stirring and heating in a solvent such as refluxing ethanol. At this stage two stereoisomers are formed. Reduction of the carbonyl radical in the presence of a suitable reducing agent, for example sodium borohydride, in a suitable solvent, such as water, alcohol, tetrahydrofuran or a mixture thereof, generally at room temperature yields the hydroxy-intermediate compound (step d), which is further converted into intermediate compound (IV') using standard techniques (step e). For example, reaction with methanesulfonylchloride or 4-methylbenzenesulfonylchloride in the presence of a base, such as triethylamine, in a reaction inert solvent, for example dichloromethane, at reaction temperatures ranging between 0°C and room temperature yields the corresponding sulfonyloxy derivative intermediate compound (IV'). The corresponding halo-derivative can also be prepared, e.g. by treating the hydroxy-intermediate compound with triphenylphosphine, in the presence of tetrachloromethane, in a reaction inert solvent, such as tetrahydrofuran, and by stirring and refluxing the mixture. In some cases a protecting group (for example a tert-butoxycarbonyl group) may be removed in step (d).

Specifically, indol-fused isoxazolidine derivatives may also be prepared as in the following reaction scheme 2

Scheme 2

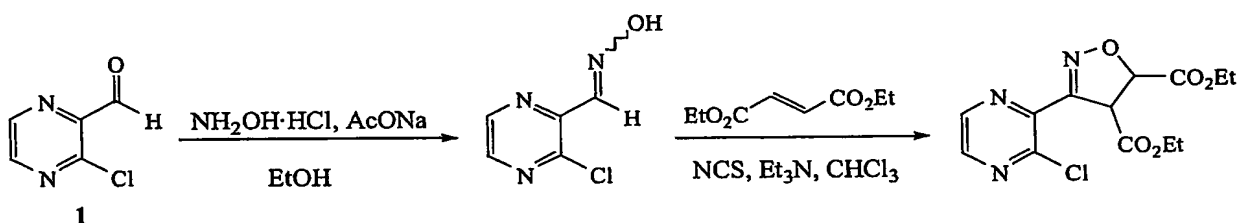
in which the hydroxyaldehyde intermediate compound is prepared as describe by Katsunori Teranishi et al. in *Synthesis*, 1994, (10), 1018-1020 by hydrolysis of the ester in the presence of a strong base, such as LiOH or NaOH, water and an inert solvent, for example 1,4-dioxane or tetrahydrofuran. The resulting hydroxy intermediate compound is formylated using art-known procedures to yield the hydroxyaldehyde

-18-

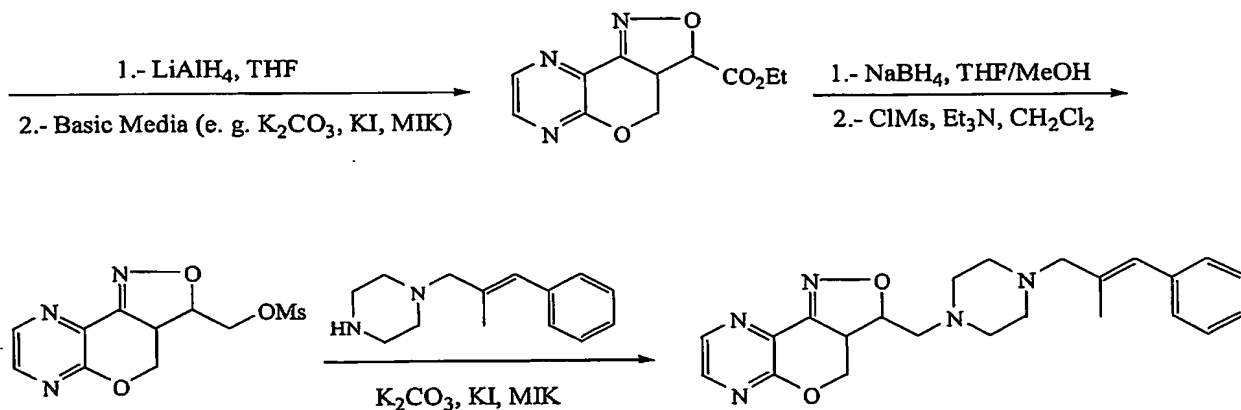
- 5 intermediate compound, such as a reaction with paraformaldehyde in the presence of an appropriate salt, for example MgCl_2 , a base, for example triethylamine or diisopropylethylamine, and in an inert solvent, for example acetonitrile or tetrahydrofuran.
- 10 Specifically, pyrazine-fused isoxazolidine derivatives may be prepared according to the following reaction scheme 3.

Scheme 3

15



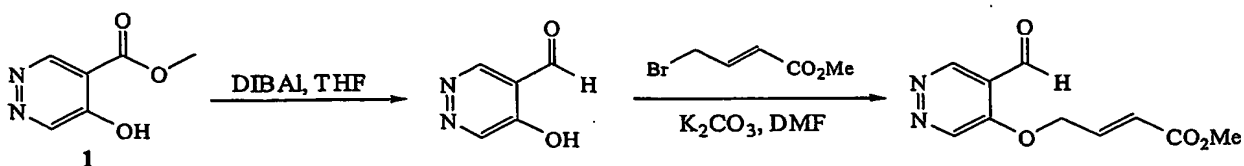
1 Described in Turck, A.; Mojovic, L.; Queguiner, G. *Synthesis*, 11, 1988, 881-884



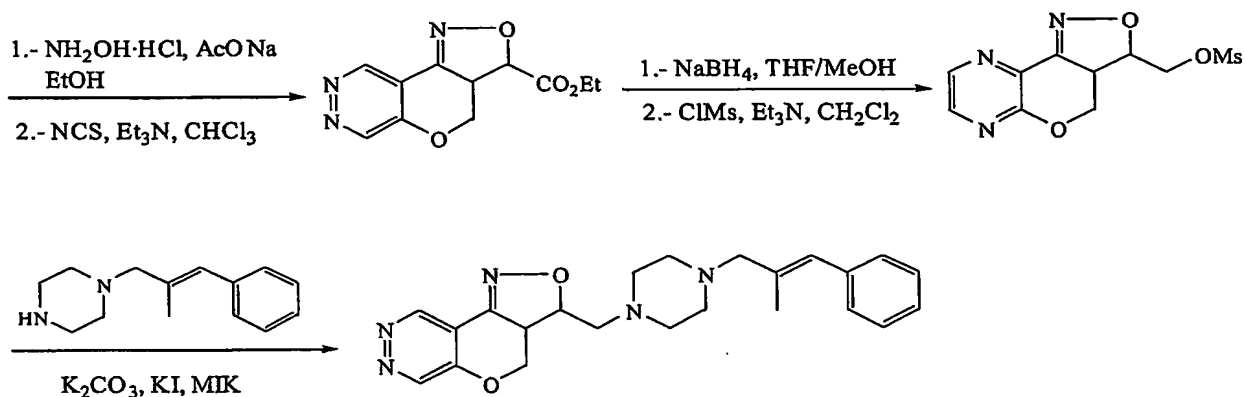
- 20 Specifically, pyridazine-fused isoxazolidine derivatives may be prepared according to either one of the following schemes 4 and 5.

-19-

Scheme 4

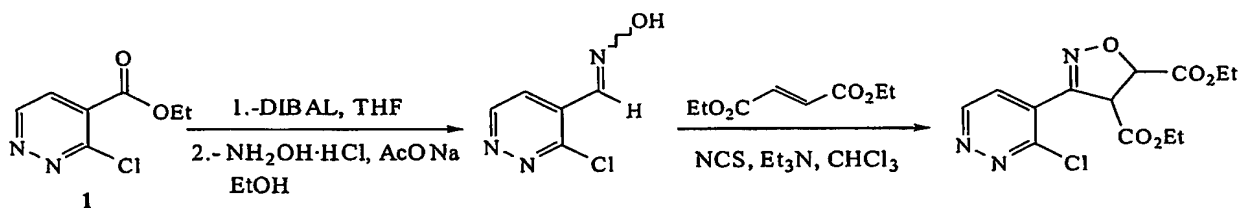


1 Described in Kinoshita, T.; Castle, R. N. J. Heterocycl. Chem., 5, 1968, 845-848

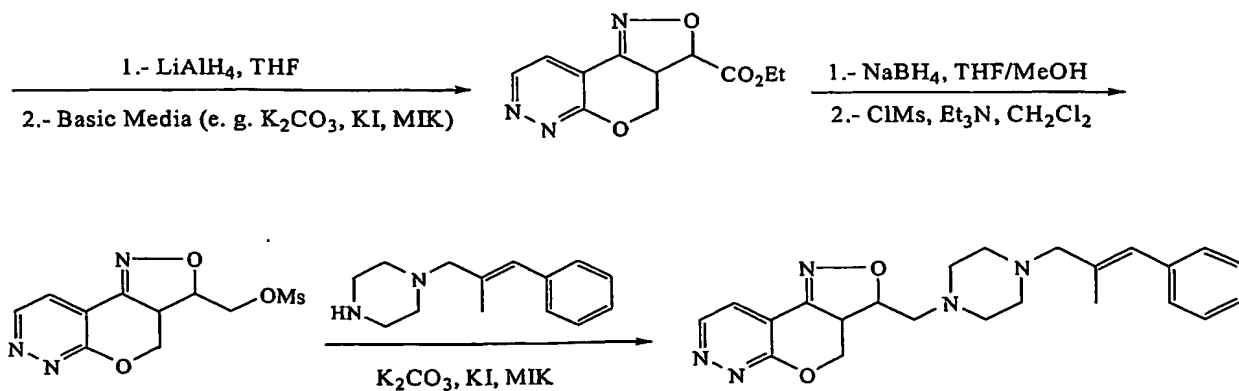


10

Scheme 5

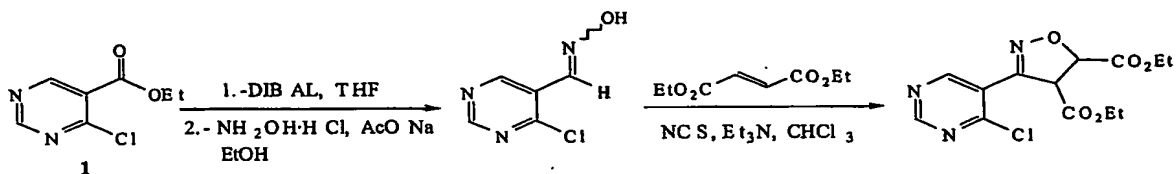


1 Described in Czech, K.; Norbert, H. G., Monatsh. Chem., 1991, 5, 413-418.
Boamah, P. Y.; Haider, N.; Heinisch, G. Arch. Pharm., 1990, 323-324.
Giani, R. P.; Malandrino, S.; Tonon, G., EP 230402 A2



-20-

- 5 Specifically, pyrimidine-fused isoxazolidine derivatives may be prepared according to either one of the following schemes 6 and 7.

Scheme 6

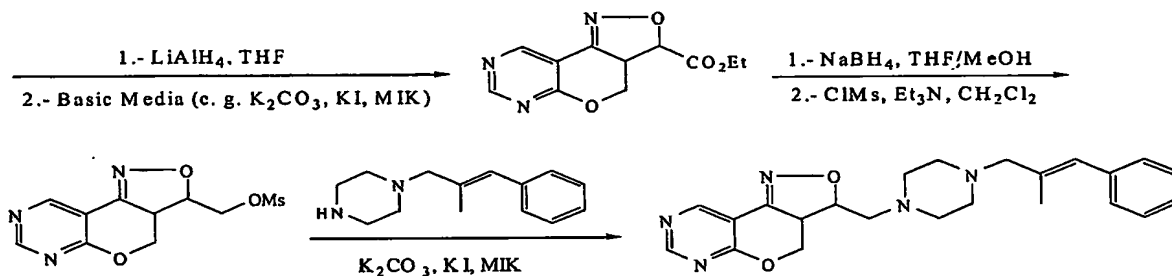
1 Described in Kubo, K.; Inada, Y.; Naka, T., CA 2106135 AA

Kubo, K.; Inada, Y.; Naka, T., EP 588299 A2

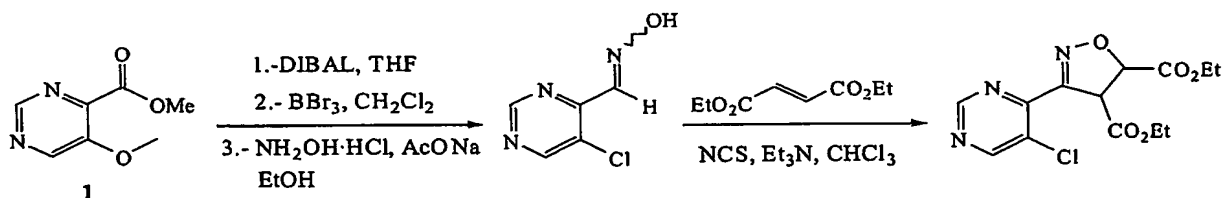
Aspisi, C.; Chiodoni, U.; Demosthene, C.; Spinelli, S., FR 2468594 A1

Pesson, M.; Antoine, M.; Chabassier, S.; Geiger, S.; Girard, P.; Richer, D.; De Lajudie, P.;

Horvath, E.; Leriche, B.; Patte, S., Eur. J. Med. Chem. - Chim. Ther., 1974, 6, 585-590.

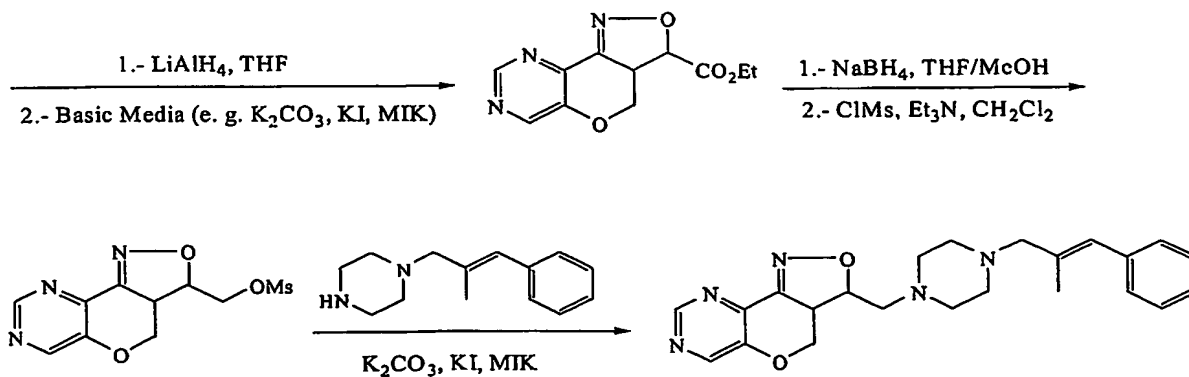


10

Scheme 7

1 Described in Barreau, M.; Cotrel, C.; Jeanmart, C., US 4110450.

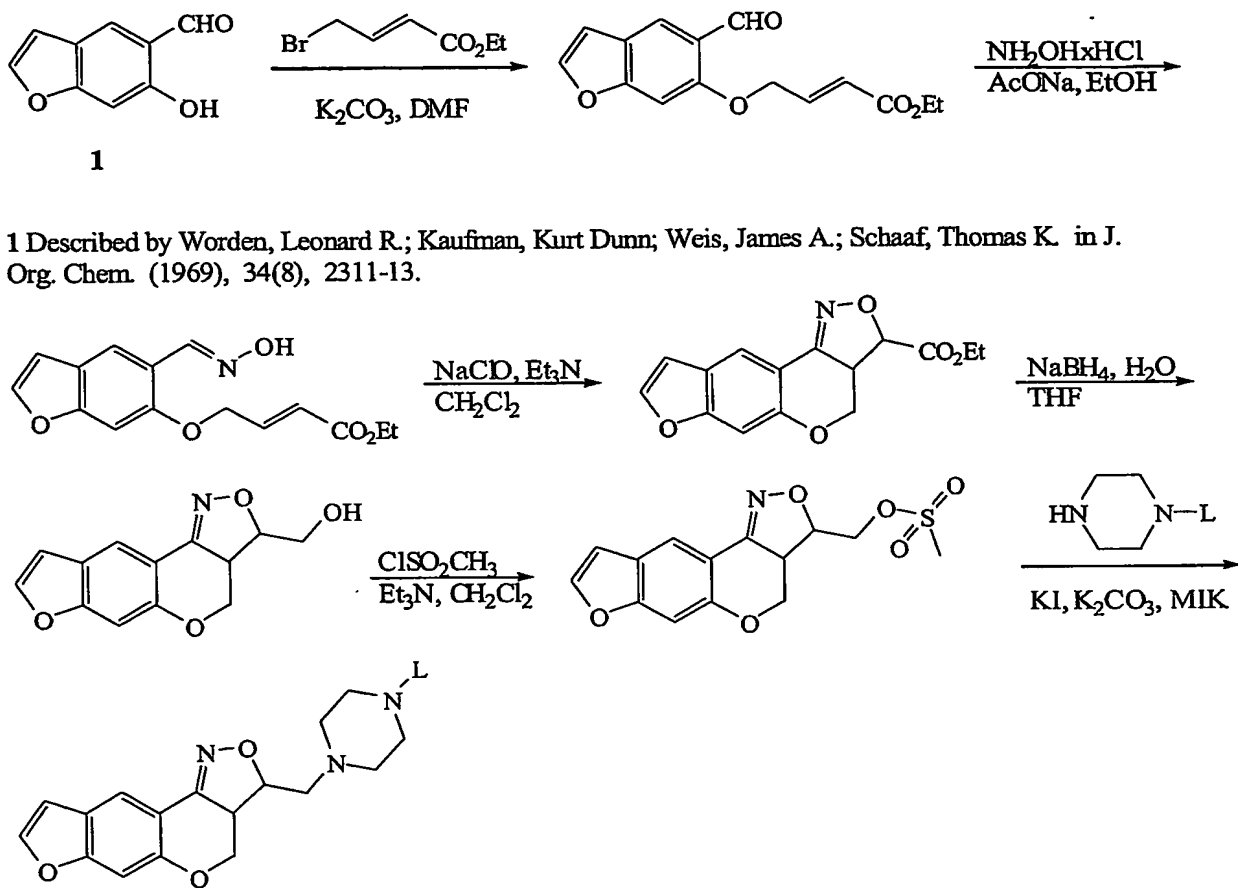
Barreau, M.; Cotrel, C.; Jeanmart, C., GE 2705641.



15

5

Specifically, benzofuran-fused isoxazolidine derivatives may be prepared according to the following scheme 8.

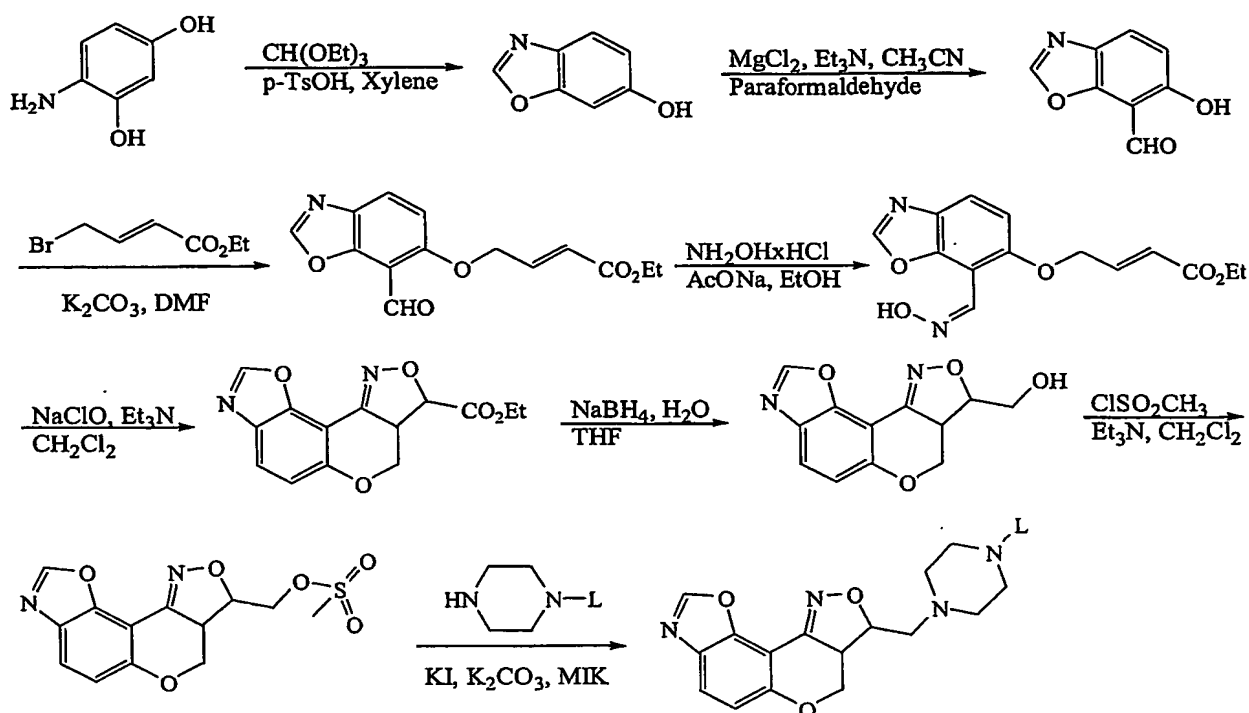
Scheme 8

15

Specifically, benzoxazole-fused isoxazolidine derivatives may be prepared according to the following scheme 9.

-22-

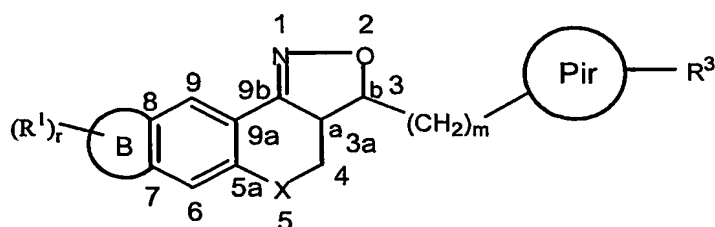
Scheme 9



10 The following examples illustrate the present invention without being limited thereto.

Experimental part

15 The carbon ring numbering system for the compounds according to Formula (I-B-b) used in this application is as follows :



20 Of some compounds the absolute stereochemical configuration of the stereogenic carbon atom(s) therein was not experimentally determined. In those cases the stereochemically isomeric form which was first isolated is designated as "A" and the second as "B", without further reference to the actual stereochemical configuration. However, said "A" and "B" isomeric forms can be unambiguously characterized by a person skilled in the art, using art-known methods such as, for example, X-ray

-23-

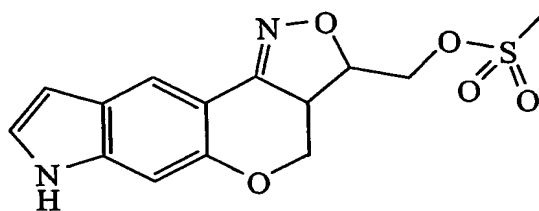
- 5 diffraction. The stereogenic centers a and b in Formula (I) have respectively the ring numbers 3a and 3.

Hereinafter, "DMF" is defined as *N,N*-dimethylformamide, "DIPE" is defined as diisopropyl ether, "ACN" is defined as acetonitrile, "DCM" is defined as
10 dichloromethane and "THF" is defined as tetrahydrofuran.

A. Preparation of the intermediate compounds

Example A.1

Preparation of intermediate compound 7



- 15 a) To a solution of 57.1g (0.176mol) of chloro-acetic acid 1-(2,2-dimethyl-propionyl)-1H-indol-6-yl ester in 500ml of 1,4-dioxane, a solution of 5.05g (0.211mol) of LiOH in 100ml of water was added portionwise at room temperature. The reaction was stirred for 1h at room temperature. then DCM and a 2N solution of HCl in water were added. The organic layer was separated, dried (Na₂SO₄), filtered off and the solvent
20 evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM and DCM/Ethyl acetate 95/5). The desired fractions were collected and the solvent evaporated. Yielding: 28.3g (74%) of 1-(6-Hydroxy-indol-1-yl)-2,2-dimethyl-propan-1-one (intermediate compound 1).
- 25 b) To a mixture of intermediate compound 1 (24.13g, 0.111mol), magnesium chloride (15.9g, 0.167mol) and diisopropylethylamine (72.5ml, 0.416mol) in 600ml of acetonitrile, 13.4g (0.443mol) of paraformaldehyde were added. the reaction was heated to reflux for 60min, then additional 13.3g (0.443mol) of paraformadehyde were added. The reaction was heated to reflux for 60min and 13.3g (0.443mol) of
30 paraformaldehyde were added again. The reaction was heated to reflux for 2h. The reaction was allowed to reach room temperature. Then a 1N solution of HCl in water and DCM were added. The organic layer was separated, dried (Na₂SO₄), filtered off and the solvent evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM). The desired fractions were collected
35 and the solvent evaporated. Yielding: 12.83g (47%) of 1-(2,2-Dimethyl-propionyl)-6-hydroxy-1H-indole-5-carbaldehyde (intermediate compound 2).

-24-

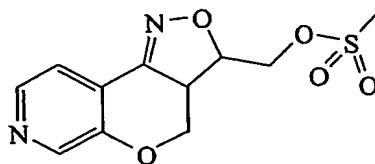
- 5 c) 4-bromo-2-butenic acid ethyl ester 14.2ml (0.082 mol) was added portionwise to a mixture of intermediate compound 2 (13.6g, 0.055 mol) and K_2CO_3 (13.68g, 0.099 mol) in DMF (60ml). The reaction mixture was stirred for 6 hours at room temperature, filtered and the filtrate was evaporated to dryness. The residue was washed with water, then extracted with DCM. The separated organic layer was dried (Na_2SO_4), filtered,
10 and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM and DCM/Ethyl acetate 9/1). The desired fractions were collected and the solvent evaporated. Yielding: 21.05 g (100%, crude yield) of 4-[1-(2,2-Dimethyl-propionyl)-5-formyl-1H-indol-6-yloxy]-but-2-enoic acid ethyl ester (intermediate compound 3).
- 15 d) Hydroxylamine hydrochloride (4.59g, 0.066 mol) was added to a mixture of intermediate compound 3 (21.05g, 0.055 mol) and sodium acetate (6.8g, 0.066mol) in ethanol (250 ml). The reaction mixture was stirred for 1h at 0°C, then water was added and extracted with DCM. The separated organic layer was dried (Na_2SO_4), filtered, and
20 the solvent was evaporated. Yielding: 21.0 g (100%, crude yield) of 4-[1-(2,2-Dimethyl-propionyl)-5-(hydroxyimino-methyl)-1H-indol-6-yloxy]-but-2-enoic acid ethyl ester (intermediate compound 4).
- e) $NaClO$, 4% (176.4ml, 0.140mol) was added portionwise at 0°C to a solution of
25 intermediate compound 4 (19.4g, 0.052 mol) in DCM (200 ml). The reaction was stirred for 1h at room temperature. Then Et_3N (10.9ml, 0.078mol) was added dropwise at 0°C. The reaction mixture was stirred for 3 hours at room temperature, then organic layer was separated, dried (Na_2SO_4), filtered, and the filtrate was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM and
30 DCM/Ethyl acetate 95/5). The desired fractions were collected and the solvent was evaporated. The residue was washed with diisopropyl ether and collected. Yielding: 4.25g (22%) of 7-(2,2-Dimethyl-propionyl)-3a,4-dihydro-3H,7H-2,5-dioxo-1,7-diaza-dicyclopenta[a,g]naphthalene-3-carboxylic acid ethyl ester (intermediate compound 5).
- 35 f) $NaBH_4$ (1.23g, 0.0325 mol) was added portionwise to a solution of intermediate compound 5 (4.25g, 0.013 mol) in THF (100 ml) and H_2O (10 ml), stirred and cooled on an ice-bath. The resulting reaction mixture was stirred for 4 hours at room temperature. The reaction mixture was treated with a 10% aqueous solution of ammonium chloride and extracted with DCM. The separated organic layer was dried
40 (Na_2SO_4), filtered and the solvent evaporated. Yielding: 3.03g of (3a,4-dihydro-3H,7H-2,5-dioxo-1,7-diaza-dicyclopenta[a,g]naphthalen-3-yl)-methanol (intermediate compound 6).

-25-

5 g) Et₃N (1.4ml, 0.0101 mol) was added to a solution of intermediate compound 6 (1.65g, 0.00675mol) in DMF (15ml). The mixture was cooled in an ice-bath. Methanesulfonyl chloride (0.58ml, 0.0075mol) was added and the resulting reaction mixture was stirred for 1h at room temperature. Then, a saturated aqueous solution of NaHCO₃ was added and the mixture was extracted with DCM, dried (Na₂SO₄), filtered and the solvent was evaporated. The residue was washed with diisopropyl ether and the solid was collected. Yielding: 1.99g (91%) of methanesulfonic acid 3a,4-dihydro-3H,7H-2,5-dioxo-1,7-diaza-dicyclopenta[a,g]naphthalen-3-ylmethyl ester (intermediate compound 7).

Example A.2

Preparation of intermediate compound 15



a) To a mixture of NaH (22.5g, 0.6mol) in 50 ml of THF a solution of 3-hydroxypyridine in 500ml of THF was added dropwise at 0°C under nitrogen atmosphere. The reaction was stirred for 15min at 0°C, then a solution of methoxymethylchloride (45ml, 0.55mol) in 200ml of THF was added portionwise at 0°C. The reaction was stirred at room temperature overnight, then quenched with a 10% aqueous solution of ammonium chloride and extracted with ethyl acetate, dried (Na₂SO₄), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Heptane/Ethyl acetate 3/1 and 2/1). The desired fraction was collected and the solvent evaporated. Yielding: 38.15g (55%) of 3-methoxymethoxy-pyridine (intermediate compound 8).

b) To a solution of intermediate compound 8 (19g, 0.14mol) and tetramethylethylenediamine (23.2ml, 0.15mol) in 500ml of ethyl ether, 60ml of a solution 2.5M of butyllithium in THF was added dropwise at -78°C under nitrogen atmosphere. The reaction was stirred for 2h at -78°C, then methyl formate (10.9ml, 0.18mol) was added dropwise at -78°C. the mixture was allowed to reach room temperature and was stirred at this temperature overnight. Then a 10% aqueous solution of citric acid was added and the organic layer was separated, dried (Na₂SO₄), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Heptane/Ethyl acetate 2/1). The desired fractions were collected and the solvent evaporated. Yielding: 18.5g (80%) of 3-methoxymethoxypyridine-4-carboxaldehyde (intermediate compound 9).

-26-

5

c) Hydroxylamine hydrochloride (17.22g, 0.248 mol) was added at 0°C to a mixture of intermediate compound 9 (34.08g, 0.208 mol) and sodium acetate (25.59g, 0.312mol) in ethanol (250ml). The reaction mixture was stirred overnight at room temperature, then water was added and extracted with DCM. The separated organic layer was dried (Na₂SO₄), filtered, and the solvent was evaporated. The residue was washed with diisopropylether with few drops of DCM and the solid was collected. Yielding: 26.24g (69%) of 3-methoxymethoxy-pyridine-4-carbaldehyde oxime (intermediate compound 10).

10

15

d) To a mixture of intermediate compound 10 (26.24g, 0.143mol) in 400ml of DCM, NaClO (4%) (485ml, 0.286mol) was added dropwise at 0°C. The reaction was stirred for 1h at 0°C and 2h at room temperature. Then dimethyl fumarate (31.0g, 0.215mol) was added at 0°C and triethylamine (29.6ml, 0.215mol) was added dropwise at 0°C. The reaction was stirred overnight at room temperature. The organic layer was separated, dried (Na₂SO₄), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Heptane/Ethyl acetate 1/1, 1/3 and pure ethyl acetate). The desired fractions were collected and the solvent evaporated. Yielding: 13.45g (29%) of 3-(3-methoxymethoxy-pyridin-4-yl)-4,5-dihydro-isoxazole-4,5-dicarboxylic acid dimethyl ester (intermediate compound 11).

20

25

e) To a solution of 13.45g (0.041mol) of intermediate compound 11, 52.1ml of a solution 1M of lithiumaluminium hydride was added dropwise at 0°C under nitrogen atmosphere. the reaction was stirred for 2h at 0°C. The excess of hydride was quenched with a saturated aqueous solution of ammonium chloride. The solid was filtered off through a CELITE pad and the filtrate was extracted with ethyl acetate. The organic layer was separated, dried (Na₂SO₄), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Ethyl acetate, Ethyl acetate/MeOH saturated with ammonia 97.5/2.5, 95/5 and 9/1). The desired fractions were collected and the solvent evaporated. Yielding: 3.8g (34%) of [4-hydroxymethyl-3-(3-methoxymethoxy-pyridin-4-yl)-4,5-dihydro-isoxazol-5-yl]-methanol (intermediate compound 12).

30

35

40

f) To a solution of intermediate compound 12 (3.8g, 0.0142mol) in 100ml of DCM, 25 ml of trifluoroacetic acid were added portionwise at room temperature. The reaction was stirred overnight at room temperature. The solvent was evaporated and the residue

-27-

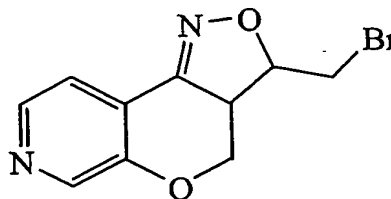
5 was co-evaporated with ethanol. Yielding 4.79g (100, crude product) of 4-(4,5-bis-hydroxymethyl-4,5-dihydro-isoxazol-3-yl)-pyridin-3-ol (intermediate compound 13).

g) To a solution of intermediate compound 13 (4.8g, 0.0142mol) in 50ml of THF, triethylamine (19.9ml, 0.0142mol) at room temperature. The mixture was stirred at
10 room temperature for 15min, then triphenyl phosphine polymer bounded (loading 1.6mmol/g) was added (17.75g, 0.0284mol) at room temperature and diethylazadicarboxylate (27.8ml, 0.0178mol) was added dropwise at room temperature. The reaction was heated to reflux for 2h. The solid was filtered off through a CELITE pad and the filtrate was evaporated. The residue was dissolved in DCM and treated
15 with water. The organic layer was separated, dried (Na_2SO_4), filtered and the solvent was evaporated. The residue was washed with acetonitrile/diisopropyl ether and the solid was collected. Yielding 3.07g (100%, crude product) of (3a,4-dihydro-3H-2,5-dioxa-1,7-diaza-cyclopenta[a]naphthalen-3-yl)-methanol (intermediate compound 14).

20 h) Intermediate compound 14 (1.72g, 0.0083mol) was treated under the reaction conditions described in Example A.1g using DCM as solvent. Yielding: 0.66g of methanesulfonic acid 3a,4-dihydro-3H-2,5-dioxa-1,7-diaza-cyclopenta[a]naphthalen-3-ylmethylester (intermediate compound 15).

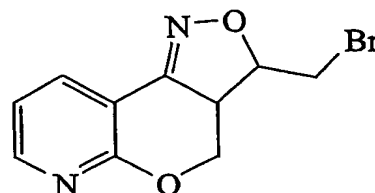
25 Example A.3

Preparation of intermediate compound 16



To a solution of intermediate compound 14 (1g, 0.0048mol) and CBr_4 (2.41g, 0.0073mol) in 100ml of DCM, triphenyl phosphine polymer bounded (loading 1.6mmol/g) was added (4.56g, 0.0073mol). the reaction was stirred overnight at room
30 temperature, then the solid was filtered off through a CELITE pad and the filtrate was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM/Ethyl acetate 1/1). The desired fractions were collected and the solvent evaporated. Yielding: 0.62g (48%) of 3-bromomethyl-3a,4-dihydro-3H-2,5-dioxa-1,7-diaza-cyclopenta[a]naphthalene (intermediate compound 16).

-28-

5 Example A.4Preparation of intermediate compound 29

a) 2-Bromopyridine-3-carboxaldehyde (9.6g, 0.052mol) was treated under the reaction conditions described in Example A.2c. Yielding: 5.91g (56%) of 2-Bromo-pyridine-3-carbaldehyde oxime (intermediate compound 17).

- 10 b) To a solution of intermediate compound 17 (5.91g, 0.0294mol) and pyridine (catalytic amount) in 100ml of CHCl_3 , *N*-chlorosuccinimide (4.32g, 0.032mol) was added portionwise at room temperature. the reaction was heated to reflux temperature for 30min. After cooling to 0°C , dimethyl fumarate (4.24g, 0.0294mol) and triethylamine (4.91ml, 0.035mol) were added. The reaction was stirred for 48h at room
- 15 temperature. Then a saturated aqueous solution of NaCO_3 was added and the organic layer was separated, dried (Na_2SO_4), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM/Ethyl acetate 95/5). The desired fractions were collected and the solvent evaporated. Yielding: 9.14g (90%) of 3-(2-bromo-pyridin-3-yl)-4,5-dihydro-isoxazole-
- 20 4,5-dicarboxylic acid dimethyl ester (intermediate compound 18).

- c) To a solution of intermediate compound 18 (9.14g, 0.027mol) in 130ml of tetrahydrofuran and 16ml of ethanol, sodium borohydride (2.51g, 0.066mol) was added portionwise at 0°C . The reaction was stirred at this temperature for 90min. Then a
- 25 saturated solution of ammonium chloride was added. the mixture was extracted with ethyl acetate and *n*-BuOH. The combined organic solutions were dried (Na_2SO_4), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Ethyl acetate, Ethyl acetate/MeOH saturated with ammonia 85/15). The desired fractions were collected and the solvent evaporated.
- 30 Yielding: 6.8g, (88%) of [3-(2-bromo-pyridin-3-yl)-5-hydroxymethyl-4,5-dihydro-isoxazol-4-yl]-methanol (intermediate compound 19).

- d) A mixture of intermediate compound 19 (6.8g, 0.0236mol), K_2CO_3 (7.2g, 0.052mol) and 18-crown-6 (1,4,7,10,13,16-hexaoxa-cyclooctadecane) (catalytic amount) in
- 35 methylisobutyl ketone was heated to reflux overnight. then water was added and the organic layer was separated, dried (Na_2SO_4), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Ethyl acetate, Ethyl acetate/MeOH saturated with ammonia 97.5/2.5, 95/5 and 9/1).

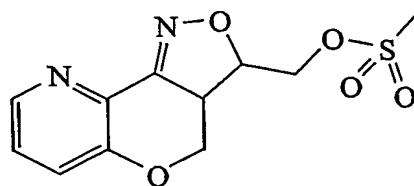
-29-

5 The desired fractions were collected and the solvent evaporated. Yielding: 0.48g (10%) of (3a,4-dihydro-3*H*-2,5-dioxo-1,6-diaza-cyclopenta[*a*]naphthalen-3-yl)-methanol (intermediate compound 20).

10 e) Intermediate compound 20 (0.46g, 0.00223mol) was treated under the reaction conditions described in Example A.3. Yielding: 0.120g (37%) of 3-bromomethyl-3a,4-dihydro-3*H*-2,5-dioxo-1,6-diaza-cyclopenta[*a*]naphthalene (intermediate compound 21).

Example A.5

Preparation of intermediate compound 29



15

a) To a solution of 2-hydroxymethyl-3-hydroxypyridine hydrochloride (10g, 0.062mol) in 100ml of DMF, imidazole (8.44g, 0.124mol) was added at room temperature. the solution was stirred for 30min at room temperature. Then tert-buthyldiphenylsilyl chloride (32ml, 0.124mol) was added dropwise at room temperature. The reaction was stirred overnight at room temperature, then the solvent was evaporated, the residue was taken up in DCM and washed with water. The organic layer was separated, dried (Na₂SO₄), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Heptane/Ethyl acetate 1/1). The desired fractions were collected and the solvent evaporated. Yielding: 12.3g (54%) of 2-(tert-butyl-diphenyl-silanyloxymethyl)-pyridin-3-ol (intermediate compound 22).

25

b) To a mixture of intermediate compound 22 (12.3g, 0.034mol) and K₂CO₃ (9.4g, 0.068mol) in DMF, 4-bromo-2-butenic acid ethyl ester (9.36ml, 0.068mol) was added dropwise at 0°C, then the reaction was stirred overnight at room temperature. The solvent was evaporated, the residue was taken up in DCM and washed with water. The organic layer was separated, dried (Na₂SO₄), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Heptane/Ethyl acetate 1/1). The desired fractions were collected and the solvent evaporated. Yielding: 8.72g (54%) of 4-[2-(tert-butyl-diphenyl-silanyloxymethyl)-pyridin-3-yloxy]-but-2-enoic acid ethyl ester (intermediate compound 23).

30

c) To a solution of pyridine (5ml) in 6ml of tetrahydrofuran, pyridinium fluoride (2.5ml) was added dropwise at 0°C under nitrogen atmosphere and the mixture was

-30-

5 stirred at that temperature for 15min. Then a solution of intermediate compound 23 (8.72g, 0.018mol) in 16ml of tetrahydrofuran was added at 0°C. The reaction was stirred at that temperature for 2h, then neutralized by addition of a saturated aqueous solution of NaHCO₃ (pH=6). The resulting mixture was extracted with DCM, the organic layer was separated, dried (Na₂SO₄), filtered and the solvent was evaporated.
10 Yielding: 7.54g (100%, crude product) of 4-(2-hydroxymethyl-pyridin-3-yloxy)-but-2-enoic acid ethyl ester (intermediate compound 24).

d) To a solution of intermediate compound 24 (7.5g, 0.032mol) in 150ml of DCM, MnO₂ (27.8g, 0.32mol) was added at room temperature. The reaction was stirred
15 overnight at room temperature, then the solid was filtered off through CELITE pad. The filtrate solvent was evaporated and the residue was purified by short open column chromatography over silica gel (eluent: Heptane/Ethyl acetate 1/1 and 1/4). The desired fractions were collected and the solvent evaporated. Yielding: 1.066g (39%) of 4-(2-formyl-pyridin-3-yloxy)-but-2-enoic acid ethyl ester (intermediate compound 25).

20 e) Intermediate compound 25 (1.066g, 0.0045mol) was treated under the reaction conditions described in Example A.1d. Yielding: 0.99g (88%) of 4-[2-(hydroxyimino-methyl)-pyridin-3-yloxy]-but-2-enoic acid ethyl ester (intermediate compound 26).

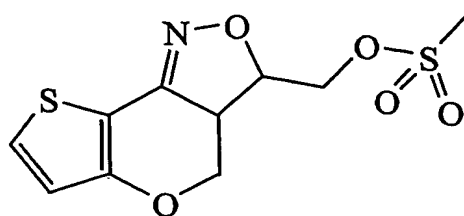
25 f) To a solution of intermediate compound 26 (0.99g, 0.00395mol) and pyridine (catalytic amount), *N*-chlorosuccinimide (0.581g, 0.00435mol) was added portionwise at room temperature and the reaction was heated to reflux for 30min. Then the reaction was cooled to room temperature and triethylamine was added (0.66ml, 0.00474mol). The reaction was stirred for 2h at room temperature, then washed with a saturated
30 aqueous solution of NaHCO₃. The organic layer was separated, dried (Na₂SO₄), filtered and the solvent was evaporated. Yielding: 0.98g (100%, crude product) of 3a,4-dihydro-3*H*-2,5-dioxo-1,9-diaza-cyclopenta[*a*]naphthalene-3-carboxylic acid ethyl ester (intermediate compound 27).

35 g) Intermediate compound 27 (0.98g, 0.0041mol) was treated under the conditions described in Example A.1f. Yielding: 0.64g (76%) of (3a,4-dihydro-3*H*-2,5-dioxo-1,9-diaza-cyclopenta[*a*]naphthalen-3-yl)-methanol (intermediate compound 28).

40 h) Intermediate compound 28 (0.64g, 0.0031mol) was treated under the conditions described in Example A.2h. Yielding: 0.702mg (80%) of methanesulfonic acid 3a,4-dihydro-3*H*-2,5-dioxo-1,9-diaza-cyclopenta[*a*]naphthalen-3-ylmethyl ester (intermediate compound 29).

-31-

5

Example A.6Preparation of intermediate compound 36

a) To a solution of 3-methoxythiophene (4.46ml, 0.043mol) in 60ml of THF, 18.92ml (0.047mol) of a 2.5M solution of butyllithium in tetrahydrofuran were added dropwise at room temperature under nitrogen atmosphere. The reaction was heated to reflux for 2h, then cooled to -10°C and DMF (4.31ml, 0.056mol) was added dropwise. the resulting reaction mixture was stirred at room temperature overnight. Then a 10% aqueous solution of ammonium chloride was added and the mixture was extracted with ethyl ether. The organic layer was separated, dried (Na_2SO_4), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM). The desired fractions were collected and the solvent evaporated. Yielding: 4.51g (73%) of 3-methoxy-thiophene-2-carbaldehyde (intermediate compound 30).

b) To a solution of intermediate compound 30 (4.51g, 0.0316mol) in 215ml of DCM, 3.3ml (0.0345mol) of BBr_3 were added at 0°C under nitrogen atmosphere. The reaction was stirred overnight at room temperature. The reaction was cooled to 0°C and additional 0.52ml (0.0055mol) of BBr_3 were added. The reaction was stirred overnight at room temperature, then treated with some drops of MeOH and with a saturated aqueous solution of ammonium chloride. The mixture was filtrated through CELITE and the filtrate organic layer was separated, dried (Na_2SO_4), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM/Ethyl acetate 96/4). The desired fractions were collected and the solvent evaporated. Yielding: 2.91g (71%) of 3-hydroxy-thiophene-2-carbaldehyde (intermediate compound 31).

c) Intermediate compound 31 (2.92g, 0.0219mol) was treated under the conditions described in Example A.1c. Yielding: 5,74g (95%) of 4-(2-formyl-thiophen-3-yloxy)-but-2-enoic acid ethyl ester (intermediate compound 32).

35

d) Intermediate compound 32 (4.87g, 0.0197mol) was treated under the conditions described in Example A.1d. Yielding: 5.78g (100%, crude product) of

-32-

- 5 4-[2-(hydroxyimino-methyl)-thiophen-3-yloxy]-but-2-enoic acid ethyl ester
(intermediate compound 33).

e) Intermediate compound 33 (5.03g, 0.0194mol) was treated under the conditions
described in Example A.1e. Yielding: 1.61g (27%) of 3a,4-dihydro-3*H*-2,5-dioxo-8-
10 thia-1-aza-as-indacene-3-carboxylic acid ethyl ester (intermediate compound 34).

f) Intermediate compound 34 (1.4g, 0.0055mol) was treated under the conditions
described in Example A.1f. Yielding: 1.02g (87%) of (3a,4-dihydro-3*H*-2,5-dioxo-8-
thia-1-aza-as-indacen-3-yl)-methanol (intermediate compound 35).

15

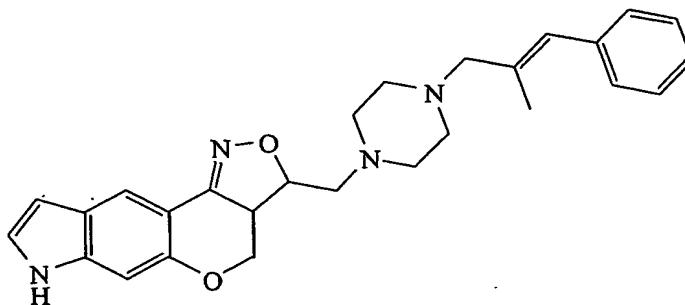
g) Intermediate compound 35 (1.17g, 0.0055mol) was treated under the conditions
described in Example A.2h. Yielding: 1.56g (97%) of methanesulfonic acid 3a,4-
dihydro-3*H*-2,5-dioxo-8-thia-1-aza-as-indacen-3-ylmethyl ester (intermediate
compound 36).

20

B. Preparation of the final compounds

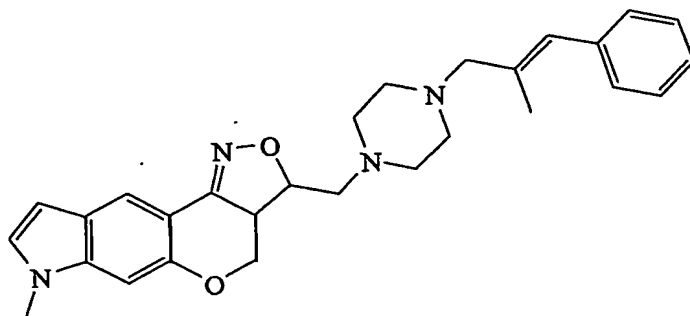
Example B.1

Preparation of final compound 1

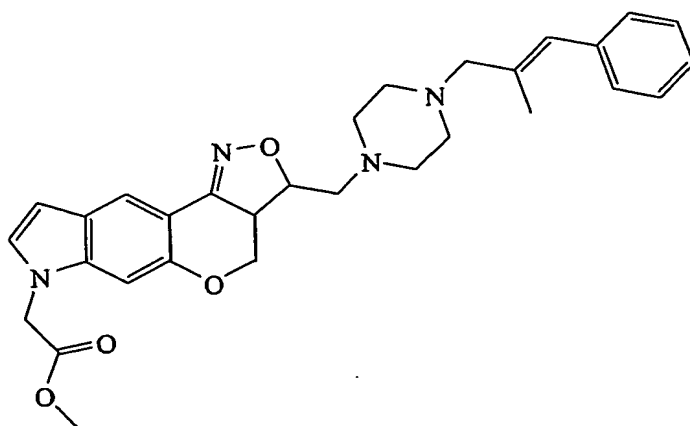


- 25 A mixture of intermediate compound 7 (prepared according to example A.1) (2.12g,
0.0066 mol) (E) 1-(2-methyl-3-phenyl-2-propenyl)piperazine (2.16g, 0.010 mol) KI
(1.1g, 0.0066mol) and K₂CO₃ (0.91g, 0.0066 mol) in methylisobutylketone (50 ml) was
stirred and refluxed overnight. Then water was added and the mixture was extracted
with DCM. The organic solution was separated, dried (MgSO₄), filtered and the solvent
30 was evaporated. The residue was purified by short open column chromatography over
silica gel (eluent: DCM/2-propanone 4/1 and 1/1). The desired fractions were collected
and the solvent evaporated. The residue was washed with diisopropylether and the solid
was collected. Yielding: 2.3g (79%) of 3-[4-(2-Methyl-3-phenyl-allyl)-piperazin-1-
ylmethyl]-3a,4-dihydro-3*H*,7*H*-2,5-dioxo-1,7-diaza-dicyclopenta[a,g]naphthalene (final
35 compound 1).

-33-

5 Example B.2Preparation of final compound 7

To a mixture of compound 1 (100mg, 0.23mmol), Na_2CO_3 (30mg, 0.28mmol), KOH
powered (16mg, 0.28mmol) in 2ml of acetonitrile and 2ml of THF; methyl iodide
(17.4 μl , 0.28mmol) was added portionwise at 0°C. The reaction was stirred at room
10 temperature overnight. Then a 10% aqueous solution of ammonium chloride was added
and the mixture was extracted with DCM. The organic solution was separated, dried
(MgSO_4), filtered and the solvent was evaporated. The residue was purified by short
open column chromatography over silica gel (eluent: DCM/MeOH 98/2). The desired
fractions were collected and the solvent evaporated. The residue was washed with
15 diisopropylether and the solid was collected. Yielding: 51.2mg (49%) of 7-methyl-3-[4-
(2-methyl-3-phenyl-allyl)-piperazin-1-ylmethyl]-3a,4-dihydro-3H,7H-2,5-dioxo-1,7-
diaza-dicyclopenta[a,g]naphthalene (final compound 7).

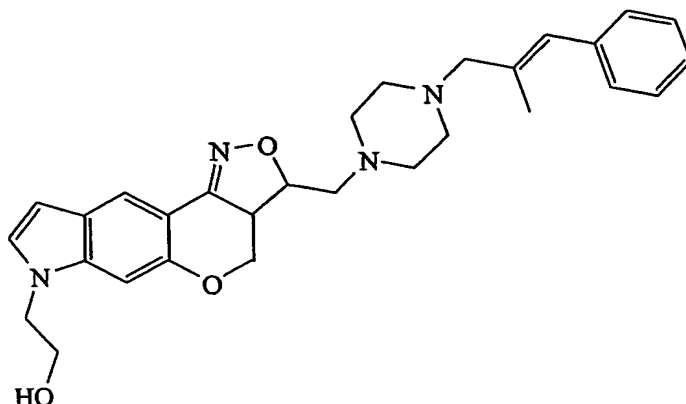
Example B.3Preparation of final compound 10

20

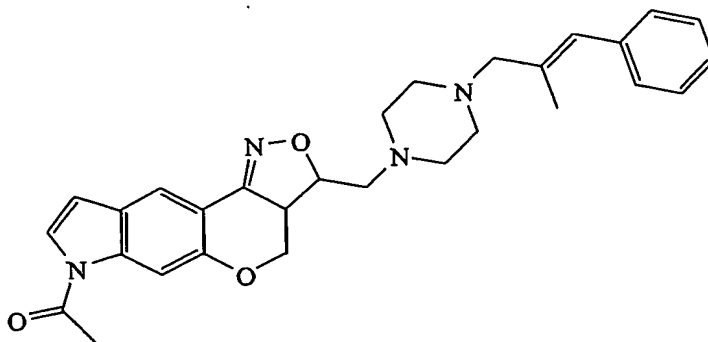
Final compound 1 (100mg, 0.23mmol) was treated with methyl bromoacetate (26.5 μl ,
0.28mmol) under the conditions described in Example B.2. Yielding: 39.0mg (33%) of
{3-[4-(2-methyl-3-phenyl-allyl)-piperazin-1-ylmethyl]-3a,4-dihydro-3H-2,5-dioxo-1,7-
diaza-dicyclopenta[a,g]naphthalen-7-yl}-acetic acid methyl ester (final compound 10).

25

-34-

5 Example B.4Preparation of final compound 8

To a solution of final compound 10 (0.24g, 4.5mmol) in 5ml of tetrahydrofuran and 1ml of MeOH, sodium borohydride (42.7mg, 1.13mmol) was added portionwise at 0°C. The reaction was stirred for 2h at room temperature. Then a 10% aqueous solution of ammonium chloride was added and the mixture was extracted with DCM. The organic solution was separated, dried (MgSO₄), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM/MeOH 98/2 and 95/5). The desired fractions were collected and the solvent evaporated. The residue was washed with diisopropylether and the solid was collected. Yielding: 87mg (40%) of 2-{3-[4-(2-methyl-3-phenyl-allyl)-piperazin-1-ylmethyl]-3a,4-dihydro-3H-2,5-dioxo-1,7-diaza-dicyclopenta[a,g]naphthalen-7-ylethanol (final compound 8) .

Example B.5Preparation of final compound 11

20

To a mixture of final compound 1 (100mg, 0.23mmol) and polymer supported BEMP (2-tert-butylimino-2-diethylamino-1,3-dimethylperhydro-1,3,2-diaza-phosphorine, loading 2.2mmol/g) (0.26g, 0.58mmol) in 2ml of DCM, acetyl chloride (40μl, 0.56mmol) was added. The reaction was stirred for 1h at room temperature. Then the solid was filtered off through a CELITE pad and the filtrate solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent:

25

-35-

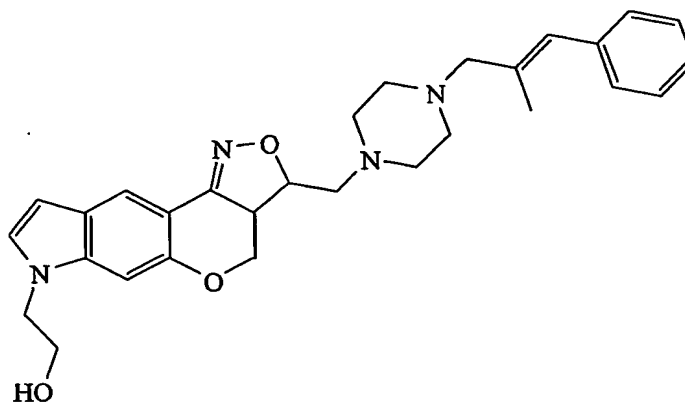
- 5 DCM/2-propanone 4/1). The desired fractions were collected and the solvent evaporated. The residue was washed with diisopropylether and the solid was collected. Yielding: 58.4mg (52%) of 1-{3-[4-(2-methyl-3-phenyl-allyl)-piperazin-1-ylmethyl]-3a,4-dihydro-3*H*-2,5-dioxo-1,7-diaza-dicyclopenta[a,g]naphthalen-7-yl}-ethanone (final compound 11).

10

Example B.6

Preparation of final compounds

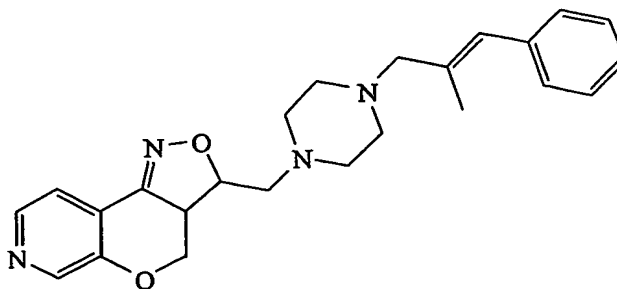
16 and 17



- Final compound 8 (prepared according to B.4) (0.8g, 0.00164mol) was purified by high-performance liquid chromatography over Chiralcel OJ (eluent: hexane/MeOH/EtOH 20/24/56). The desired fractions were collected and the solvent was evaporated. Yield: fractions A and B.
- 15 Fraction A: 256mg (32%) of A-2-{3-[4-(2-methyl-3-phenyl-allyl)-piperazin-1-ylmethyl]-3a,4-dihydro-3*H*-2,5-dioxo-1,7-diaza-dicyclopenta[a,g]naphthalen-7-yl}-ethanol (final compound 16).
- 20 Fraction B: 276mg (35%) of B-2-{3-[4-(2-methyl-3-phenyl-allyl)-piperazin-1-ylmethyl]-3a,4-dihydro-3*H*-2,5-dioxo-1,7-diaza-dicyclopenta[a,g]naphthalen-7-yl}-ethanol (final compound 17).

Example B.7

Preparation of final compound 18



25

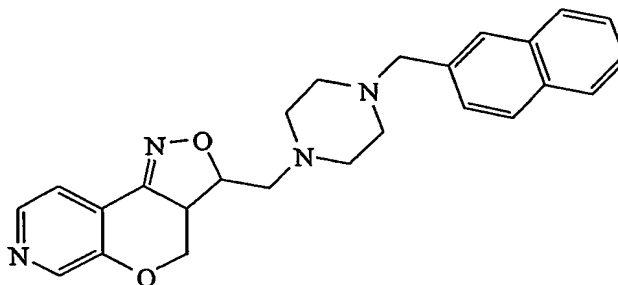
A mixture of intermediate compound 16 (prepared according to example A.3) (0.15g, 0.56mmol) and (E) 1-(2-methyl-3-phenyl-2-propenyl)piperazine (0.242g, 1.12mmol) in

-36-

- 5 1ml of 1,4-dioxane was heated at 100°C for 1h. The mixture was taken up in DCM and washed with water. The organic solution was separated, dried (MgSO₄), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: DCM/Ethyl acetate 1/1, 1/2 and pure Ethyl acetate). The desired fractions were collected and the solvent evaporated. The residue
10 was converted into its etanodioic acid salt in EtOH and the solid was collected. Yielding: 76mg (27%) of 3-[4-(2-methyl-3-phenyl-allyl)-piperazin-1-ylmethyl]-3a,4-dihydro-3H-2,5-dioxo-1,7-diaza-cyclopenta[a]naphthalene oxalate salt (1/1) (final compound 18).

15 Example B.8

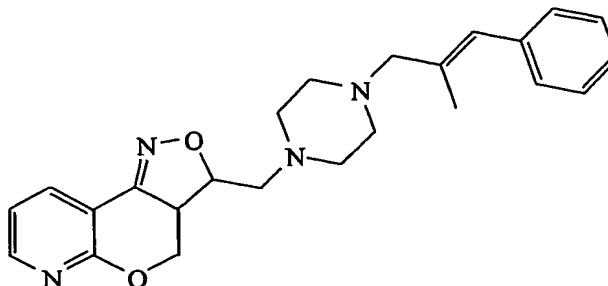
Preparation of final compound 19



- Intermediate compound 15 (prepared according to example A.2) (72mg, 0.253mmol) was treated with 1-Naphthalen-2-ylmethyl-piperazine (63mg, 0.278mmol) under the conditions described in Example B.1. The product was converted into its ethanodioic acid salt in EtOH. The solvent was evaporated and the residue was washed with
20 acetonitrile/diisopropyl ether. The solid was collected. Yielding: 11mg (8%) of 3-(4-naphthalen-2-ylmethyl-piperazin-1-ylmethyl)-3a,4-dihydro-3H-2,5-dioxo-1,7-diaza-cyclopenta[a]naphthalene oxalate salt (1/1) (final compound 19).

25 Example B.9

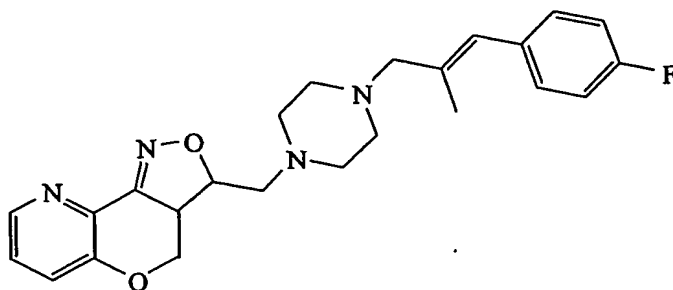
Preparation of final compound 22



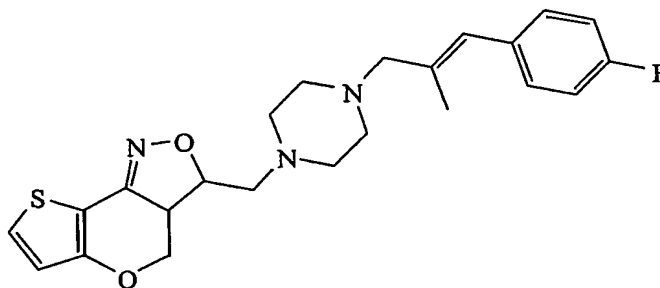
- Intermediate compound 21 (prepared according to example A.4) (120mg, 0.446mmol) and (E) 1-(2-methyl-3-phenyl-2-propenyl)piperazine (192mg, 0.892mmol) in 1ml of 1,4-dioxane was treated under the conditions described in Example B.7. Yielding:
34mg (15%) of 3-[4-(2-methyl-3-phenyl-allyl)-piperazin-1-ylmethyl]-3a,4-dihydro-3H-
30 2,5-dioxo-1,6-diaza-cyclopenta[a]naphthalene oxalate salt (final compound 22).

-37-

5

Example B.10Preparation of final compound 24

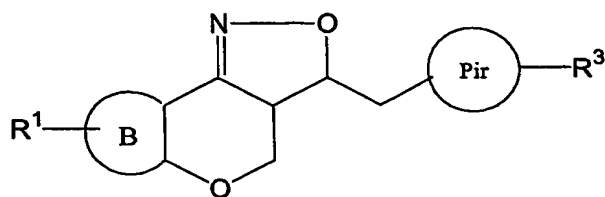
A mixture of intermediate compound 29 (prepared according to example A.5) (200mg, 0.703mmol), 1-[3-(4-Fluoro-phenyl)-2-methyl-allyl]-piperazine (198mg, 0.844mmol) and K_2CO_3 (117g, 0.844mmol) in methylisobutylketone (5 ml) was stirred and refluxed
10 overnight. Then water was added and the mixture was extracted with DCM. The organic solution was separated, dried ($MgSO_4$), filtered and the solvent was evaporated. The residue was purified by short open column chromatography over silica gel (eluent: Heptane/Ethyl acetate 3/7, 2/8 and Ethyl acetate). The desired fractions were collected and the solvent evaporated. The residue was washed with acetonitrile/diisopropylether
15 and the solid was collected. Yielding: 89mg (30%) of 3-{4-[3-(4-fluoro-phenyl)-2-methyl-allyl]-piperazin-1-ylmethyl}-3a,4-dihydro-3H-2,5-dioxo-1,9-diazacyclopenta[a]naphthalene (final compound 24).

Example B.11Preparation of final compound 27

20 Intermediate compound 36 (prepared according to example A.6) (0.5g, 0.0017mol) and 1-[3-(4-Fluoro-phenyl)-2-methyl-allyl]-piperazine (0.81g, 0.0034mol) were treated under the conditions described in Example B.1. Yielding: 0.15g (20%) of 3-{4-[3-(4-fluoro-phenyl)-2-methyl-allyl]-piperazin-1-ylmethyl}-3a,4-dihydro-3H-2,5-dioxo-8-thia-1-aza-as-indacene (final compound 27).

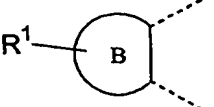
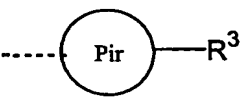
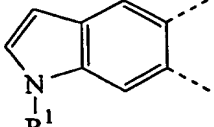
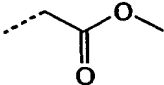
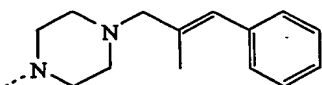
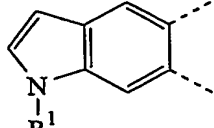

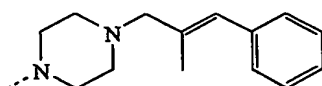
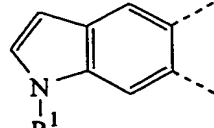
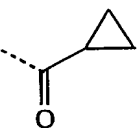
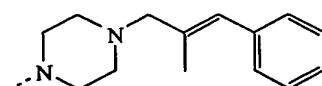
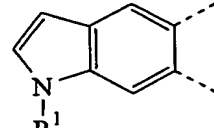
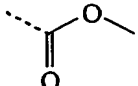
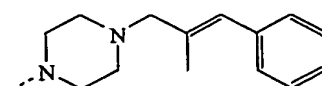
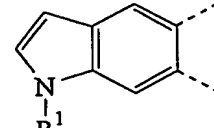
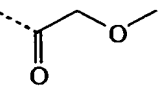
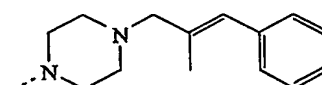
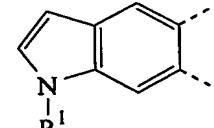

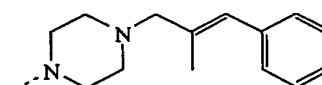
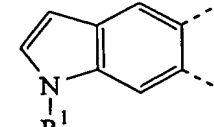
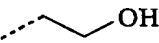
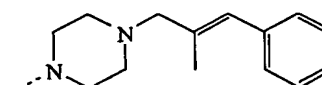
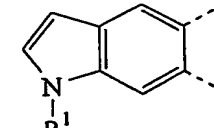
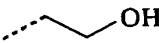
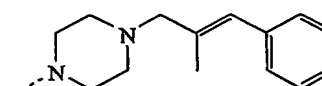
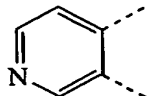
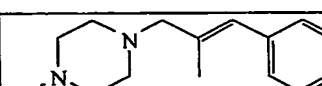
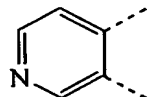
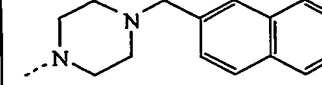
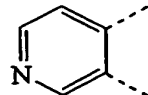
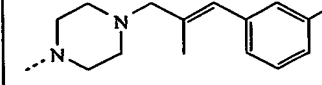
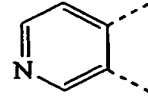
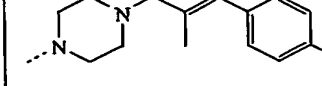
25

The following final compounds were made accordingly :

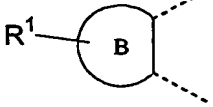
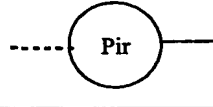
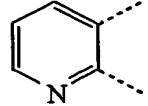
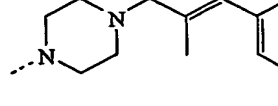
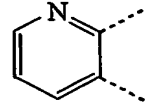
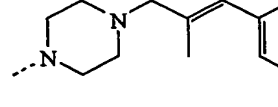
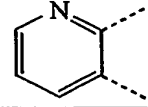
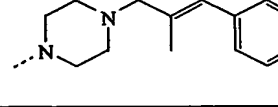
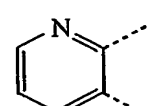
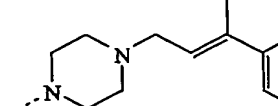
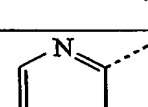
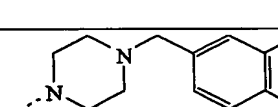
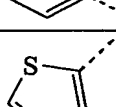
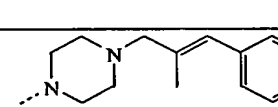
5 Table 1

Co. No.	Exp No.	R^1 -B	$-R^1$	Pir- R^3	Phys.data
1	B.1		-H		[3 α (E),3 $\alpha\alpha$]
2	B.1		-H		[3 α (E),3 $\alpha\alpha$]
3	B.1		-H		[3 α (E),3 $\alpha\alpha$]
4	B.1		-H		[3 α ,3 $\alpha\alpha$]
5	B.1		-H		[3 α ,3 $\alpha\alpha$]
6	B.1		-H		[3 α (E),3 $\alpha\alpha$]
7	B.2		-CH ₃		[3 α (E),3 $\alpha\alpha$]
8	B.4				[3 α (E),3 $\alpha\alpha$]
9	B.2				[3 α (E),3 $\alpha\alpha$]

-39-

Co. No.	Exp No.	R^1 	$-R^1$	 R^3	Phys.data
10	B.3				[3 α (E),3 $\alpha\alpha$]
11	B.5				[3 α (E),3 $\alpha\alpha$]
12	B.5				[3 α (E),3 $\alpha\alpha$]
13	B.5				[3 α (E),3 $\alpha\alpha$]
14	B.5				[3 α (E),3 $\alpha\alpha$]
15	B.2				[3 α (E),3 $\alpha\alpha$]
16	B.6				A- [3 α (E),3 $\alpha\alpha$]
17	B.6				B- [3 α (E),3 $\alpha\alpha$]
18	B.7		----		[3 α (E),3 $\alpha\alpha$] C ₂ H ₂ O ₄ (1:1)
19	B.8		----		[3 α ,3 $\alpha\alpha$] C ₂ H ₂ O ₄ (1:1)
20	B.8		----		[3 α (E),3 $\alpha\alpha$]
21	B.7		----		[3 α (E),3 $\alpha\alpha$] C ₂ H ₂ O ₄ (1:1)

-40-

Co. No.	Exp No.	R ¹ 	-R ¹	 R ³	Phys.data
22	B.9		----		[3α(E),3αα] C ₂ H ₂ O ₄ (1:1)
23	B.10		----		[3α(E),3αα] C ₂ H ₂ O ₄ (1:2)
24	B.10		----		[3α(E),3αα]
25	B.10		----		[3α(E),3αα]
26	B.10		----		[3α,3αα]
27	B.11		----		[3α(E),3αα]

5

For a selection of compounds, melting points were obtained with a Büchi melting point apparatus B-545. The heating medium is a metal block. The melting of the sample is visually observed by a magnifying lense and a big light contrast. Melting points are measured with a temperature gradient of 3 degrees Celsius/minute. The results are summarized in Table 1b.

10

Table 1b : Melting points

Co. No.	Melting point (°C)	Visual observation
1	173.2-179.1	At 173.2°C shrink, at 177.1°C red foam crystals, at 179.1°C red liquid
2	169.8-184.3	At 169.8°C shrink, at 181.5°C red foam crystals, at 184.3 red liquid
3	158.9-170.1	At 158.9°C shrink, at 170.1°C red brown liquid
4	176.6-207.3	At 176.6°C shrink, at 207.3°C red sticky product
5	131.1-154.6	At 131.1°C shrink, at 154.6°C brown liquid
6	91.6-99.2	At 91.6°C shrink, at 99.2°C colourless liquid and orange sticky

-41-

Co. No.	Melting point (°C)	Visual observation
12	107.5-114.2	At 107.5°C shrink, at 114.2°C yellow liquid
16	167.9-170.2	At 167.9°C shrink, at 170.2°C light yellow liquid
17	167.5-170.5	At 167.5°C shrink, at 170.5°C yellow liquid
18	216.8-220.5	At 216.8°C shrink, at 218.4°C black foam crystals, at 220.5°C black liquid
20	128.1-136.8	At 128.1°C shrink, at 136.8°C light yellow liquid
24	145.6-152.3	At 145.6°C shrink, at 152.3°C black liquid
25	128.1-131.1	At 128.1°C shrink, at 131.1°C light yellow liquid
26	152.4-158.1	At 152.4°C shrink, at 158.4°C light yellow liquid
27	142.3-146.0	At 142.3°C shrink, at 146.0°C colourless liquid

5

C. Pharmacological examplesExample C1 : Binding experiment for α_2 -adrenergic receptor subtypes and for 5-HT transporter

10

General

The interaction of the compounds of Formula (I) with α_2 -receptors and h5-HT-transporters was assessed in *in vitro* radioligand binding experiments. In general, a low concentration of a radioligand with a high binding affinity for a particular receptor or transporter is incubated with a sample of a tissue preparation enriched in a particular receptor or transporter or with a preparation of cells expressing cloned human receptors in a buffered medium. During the incubation, the radioligand binds to the receptor or transporter. When equilibrium of binding is reached, the receptor bound radioactivity is separated from the non-bound radioactivity, and the receptor- or transporter-bound activity is counted. The interaction of the test compounds with the receptor is assessed in competition binding experiments. Various concentrations of the test compound are added to the incubation mixture containing the receptor- or transporter preparation and the radioligand. The test compound in proportion to its binding affinity and its concentration inhibits binding of the radioligand. The radioligand used for α_2A , α_2B and α_2C receptor binding was [3H]-rauwolscine and for the h5-HT transporter was [3H]paroxetine.

25

-42-

5 Cell culture and membrane preparation.

CHO cells, stably transfected with human adrenergic- α_{2A} -, - α_{2B} or α_{2C} receptor cDNA, were cultured in Dulbecco's Modified Eagle's Medium (DMEM)/Nutrient mixture Ham's F12 (ratio 1:1)(Gibco, Gent-Belgium) supplemented with 10 % heat inactivated fetal calf serum (Life Technologies, Merelbeke-Belgium) and antibiotics (100 IU/ml penicillin G, 100 μ g/ml streptomycin sulphate, 110 μ g/ml pyruvic acid and 100 μ g/ml L-glutamine). One day before collection, cells were induced with 5 mM sodiumbutyrate. Upon 80-90 % of confluence, cells were scraped in phosphate buffered saline without Ca^{2+} and Mg^{2+} and collected by centrifugation at 1500 x g for 10 min. The cells were homogenised in Tris-HCl 50 mM using an Ultraturrax homogenizer and centrifuged for 10 min at 23,500 x g. The pellet was washed once by resuspension and rehomogenization and the final pellet was resuspended in Tris-HCl, divided in 1 ml aliquots and stored at -70°C .

Binding experiment for α_2 -adrenergic receptor subtypes

20 Membranes were thawed and re-homogenized in incubation buffer (glycylglycine 25 mM, pH 8.0). In a total volume of 500 μ l, 2-10 μ g protein was incubated with [^3H]rauwolscine (NET-722) (New England Nuclear, USA) (1 nM final concentration) with or without competitor for 60 min at 25°C followed by rapid filtration over GF/B filter using a Filtermate196 harvester (Packard, Meriden, CT). Filters were rinsed extensively with ice-cold rinsing buffer (Tris-HCl 50 mM pH 7.4). Filter-bound radioactivity was determined by scintillation counting in a Topcount (Packard, Meriden, CT) and results were expressed as counts per minute (cpm). Non-specific binding was determined in the presence of 1 μ M oxymetazoline for $\text{h}\alpha_{2A}$ - and $\text{h}\alpha_{2B}$ receptors and 1 μ M spiroxatrine for $\text{h}\alpha_{2C}$ receptors.

30

Binding experiment for 5-HT transporter

Human platelet membranes (Oceanix Biosciences Corporation, Hanover, MD, USA) were thawed, diluted in buffer (Tris-HCl 50 mM, 120 mM NaCl and 5 mM KCl) and quickly (max 3 s) homogenised with an Ultraturrax homogenizer. In a total volume of 250 μ L, 50-100 μ g protein was incubated with [^3H]paroxetine (NET-869) (New England Nuclear, USA) (0.5 nM final concentration) with or without competitor for 60 min at 25°C . Incubation was stopped by rapid filtration of the incubation mixture over GF/B filters, pre-wetted with 0.1 % polyethylenamine, using a Filtermate196 harvester (Packard, Meriden, CT). Filters were rinsed extensively with ice-cold buffer and radioactivity on the filters was counted in a Topcount liquid scintillation counter (Packard, Meriden, CT). Data were expressed as cpm. Imipramine (at 1 μ M final concentration) was used to determine the non-specific binding.

5

Data analysis and results

Data from assays in the presence of compound were calculated as a percentage of total binding measured in the absence of test compound. Inhibition curves, plotting percent of total binding versus the log value of the concentration of the test compound, were automatically generated, and sigmoidal inhibition curves were fitted using non-linear regression. The pIC₅₀ values of test compounds were derived from individual curves.

All compounds according to Formula (I) produced an inhibition at least at the hα_{2A} site (but often also at the hα_{2B} and hα_{2C} sites) and simultaneously at the 5-HT transporter site of more than 50 % (pIC₅₀) at a test concentration ranging between 10⁻⁶ M and 10⁻⁹ M in a concentration-dependent manner.

Table 2 : Pharmacological data.

Co. No	hα _{2A}	hα _{2B}	hα _{2C}	5-HTT
20	9.2	-	8.8	7.6
18	8.9	-	8.8	6.7
23	8.9	-	8.4	6.7
18	8.8	8.8	8.8	7.5
27	8.7	-	8.1	7.1
25	8.7	-	7.8	6.2
11	8.6	-	9.0	8.2
14	8.5	-	8.9	8.4
7	8.5	-	9.1	8.3
13	8.4	-	8.4	7.9
1	8.3	8.4	9.0	8.4
24	8.2	-	7.5	6.7
21	8.2	-	7.8	7.1
12	8.2	-	8.5	7.7
26	8.1	-	7.3	7.4
6	8.1	-	8.6	8.3
2	8.1	8.3	9.0	8.3
9	8.1	8.1	9.1	8.7
19	8.0	-	7.4	7.0
17	8.0	8.3	8.7	6.3
10	7.9	8.3	9.1	8.7
15	7.9	8.4	8.6	8.3

-44-

Co. No	h α 2A	h α 2B	h α 2C	5-HTT
3	7.8	7.6	8.7	8.2
8	7.8	8.3	8.7	8.4
4	7.6	7.3	8.4	8.8
5	6.7	-	7.4	7.8
16	6.5	7.7	7.3	6.3
22	6.2	-	7.1	5.2